Status of Program

The Agricultural Research Services' (ARS) major research programs -- New Products/Product Quality/Value Added; Livestock/Crop Production; Food Safety; Livestock/Crop Protection; Human Nutrition; and Environmental Stewardship -- address the Department's goals and priorities. A brief summary of the agency's selected accomplishments and current research activities including the National Agricultural Library are detailed below.

New Products/Product Quality/Value Added (ARS Goal 2) (306, 213)

Select Examples of Recent Progress:

NP306

Starch-oil composite gel fat replacer for ground meat applications. This technology was developed by scientists at Peoria, Illinois and commercialized resulting in large scale production of a fat replacer gel and reduced fat ground beef patties. Use of the gel enables the conversion of 93 to 95 percent lean beef, which tends to yield a chewy and dry cooked patty, to a low fat alternative which consistently provides tenderness, juiciness, and flavor (delivered in the lipid phase of the gel). Similar results of this technology were also demonstrated in meatballs and pork sausages. The company plans to expand the marketing of the low fat patties to large volume institutional customers as well as broaden the product line to include emulsified meat products, such as frankfurters and lunch meats. The availability of this technology, which delivers significant fat (and calorie) reduction while enhancing product quality, will ultimately enable progress in addressing consumer obesity, and increasing acceptance of healthier alternatives to traditionally high fat foods. (NP 306, C 2, P.S. 2a and 2c, P.M. 2.1.2)

Commercialization of estolides as a biobased lubricant. There is a demand in the U.S. and worldwide for increased development and use of biobased products. Scientists at Peoria, Illinois developed and patented biobased lubricants called "estolides" as biobased lubricants. A private sector partner has advanced commercialization, production, and availability of the product to future customer/industry users. The first commercial production will take place in early FY 2011. Development of estolides will lessen U.S. demand on foreign oil while decreasing the amounts of petroleum-based pollutants being released into the environment. (NP 306, C 2, P.S. 2a and 2c, P.M. 2.1.2)

Adapting NIRS grain technology for detecting traits of disease vectors. Engineers in Manhattan, Kansas developed a technology to measure traits of single wheat kernels using near-infrared spectroscopy (NIRS). This technology can also determine traits of single insects, such as their species and age. In cooperative work with the Centers for Disease Control (CDC) in Atlanta, Georgia and the Ifakara Health Institute in Ifakara, Tanzania, researchers demonstrated that the NIRS technology can determine mosquito species and age with about 90 percent accuracy. These findings are important in monitoring control programs and reducing the proportion of older mosquitoes that have the ability to transmit malaria. The technology has been adopted by the CDC, and researchers in England, Austria, Australia, and Tanzania. (NP 306, C 1, P.S. 1b, P.M. 2.1.2)

<u>Development of improved wheat germplasm</u>. Wheat producers, milling and baking industries, and overseas customers require high standards in the quality of wheat to meet their evolving needs. Scientists in Fargo, ND, contributed wheat end-use quality data that helped lead to the development of improved wheat germplasm and subsequent release of new cultivars of spring, winter, and durum wheat bred for commercial production. The lab provided over 40 different tests related to the physical and biochemical quality traits of the wheat kernel and related milling performance, flour, semolina, dough, baking, and

spaghetti processing on over 4000 samples of hard spring, hard winter, and durum wheat lines that were submitted by public and private wheat breeders and cooperating scientists. The impact lies in the release of 5 experimental lines of spring wheat, which included the cultivar Brick. (NP 306, C 1, P.S. 1a, 1b, and 1c, P.M. 2.1.2)

Ozone fumigation controls postharvest decay of table grapes. Table grapes rot and spoil after harvest unless actions are taken to preserve them, such as the fumigation of grape storage rooms with sulfur dioxide; however, growers of 'organic' grapes cannot use sulfur dioxide and need an acceptable option. Researchers in Parlier, CA, evaluated ozone fumigation in Parlier and in two commercial cold storages. Ozone gas, an approved substance for use by 'organic' growers under the USDA National Organic Program rules, reduced rot and approximately doubled the storage life of the grapes in cold storage from 2-3 weeks to 4-5 weeks. This work provides a useful alternative method to reduce rot and extend the storage life of grapes in this industry, which produced 99 million boxes worth approximately \$1,200,000,000 in California in 2008, of which approximately 5% were classified as 'organic'. (NP 306, C 1, P.S. 1d, P.M. 2.1.2)

<u>Use of cotton byproducts in bio-materials</u>. Expanded Polystyrene (EPS) and Expanded Polypropylene (EPP) are used to make non-biodegradable protective packaging materials, a \$2.2 billion industry. Engineers at Lubbock, TX, developed six cotton byproduct blends and mechanical processing techniques that allowed for the cooperator to manufacture 100% biodegradable packaging composites, using their technology, which outperformed EPS and EPP. The cotton byproduct protective packaging material resulting from this research allowed the cooperator to launch the product in 2010 with a Fortune 500 company and gain the interest of two other Fortune 500 companies. More widespread use of cotton byproducts is expected within the next few years. (NP 306, C 2, P.S. 2a and 2c, P.M. 2.1.2)

Biobased materials in tires. Researchers in Albany, CA, in collaboration with Cooper Tire and Rubber and the National Renewable Energy Laboratory, conducted a Life Cycle Analysis comparing petroleum-based and bio-based material usage in tire manufacturing. The analysis estimated the potential impact of biobased materials technology on reducing oil dependency related to 1) raw material manufacture, 2) gasoline savings from improved fuel efficiency, 3) net energy savings associated in making raw materials, and 4) the potential impact on greenhouse gas emissions. The use of natural rubber in all tire components was premised. The analysis effectively demonstrated the positive contributions the U.S. tire industry could make toward the goal of decreasing petroleum dependency by converting from petroleum based materials to non-petroleum based materials. (NP 306, C 2, P.S. 2a, P.M. 2.1.2)

Commercialization of sorting technology and adoption by seed breeders/producers. Engineers at Manhattan, KS, commercialized and transferred a low cost color image based sorting device for grains to National Manufacturing through a CRADA. These instruments have been sold to various seed breeders and seed foundations in the US and internationally. The new sorting system has unprecedented accuracy, throughput, and low cost for inspection/sorting systems. A CRADA with Jolly Time Popcorn is also in place to adapt the machine for sorting popcorn, and the camera design has been transferred to an electronics manufacturer (Short Dog Electronics). These instruments have been extensively used for separation of large bulks of popcorn, yellow and brown flax, red and white wheat, scab-damaged wheat, and removing weed seeds from seed stocks to improve quality of breeding lines and end-use products. North Dakota State University seed foundation states that the machines shortened production time for yellow flax by one year, increased production by 20%, and reduced contaminates by 90% over past practices. Other users report similar impact. (NP 306, C 1, P.S. 1b, P.M. 2.1.2)

<u>Moisture sensing in almond kernels</u>. For almonds, initial moisture content and moisture migration are critical for safe storage and the preservation of their quality attributes, and there is a need for real-time monitoring of moisture content at different stages of processing. At the request of the Almond Board of

California, engineers at Athens, GA, adapted a microwave dielectric method, which was originally developed for grains and seeds, to rapidly predict moisture content in almond kernels. Adoption of this technology by almond growers and processors will allow them to improve the quality of almonds, avoid spoilage and waste, and maintain their position as the global market leader. (NP 306, C 1, P.S. 1b, P.M. 2.1.2)

<u>Pre-harvest sprays reduce citrus postharvest diseases</u>. Canker and melanose diseases cause postharvest rejection of citrus fruit. Pre-harvest sprays of a wax emulsion combined with pesticide, applied by scientists from Winter Haven, FL, along with industry cooperators, was successful in reducing canker bacteria and melanose fungus on citrus leaves and fruit in the field, resulting in less inoculum when the fruit entered the packing house and less postharvest decay and eliminations. The reason for the reduced pathogen load due to the pre-harvest treatment was because the wax spray allowed the pesticide to remain longer on the tree during rain events. This process reduces postharvest problems and resulted in a patent application. (NP 306, C 1, P.S. 1d, P.M. 2.1.2)

Commercialization of pterostilbene. Pterostilbene is a naturally-occurring phenolic compound in blueberries and an analog of resveratrol which is the well-known polyphenol in grapes and wine. Scientists at Oxford, MS, showed that pterostilbene is a more effective cholesterol lowering agent than resveratrol in laboratory animals, as far as activating a protein that plays a major role in lipid metabolism and transport. In another animal study, pterostilbene was shown to reverse memory and cognition deficits. For these activities and the potential to explore similar effects in humans, pterostilbene was licensed by a company (ChromaDex). Commercialization of pterostilbene under the tradename pTeroPureTM was launched in April 2010. pTeroPureTM pterostilbene is now on the market for use as a pure compound, or mixed with other natural compounds as dietary supplements. More pterostilbene products are expected to be on the market in the coming years. (NP 306, C 2, P.S. 2a, P.M. 2.1.2)

Effect of Huanglongbing disease on orange juice flavor. Huanglongbing (HLB), also known as 'greening', is a devastating disease for the citrus industry. In addition to debilitating trees, fruit quality was anecdotally reported to be affected, but no chemical or sensory studies had been done. Researchers in Winter Haven, FL, showed that only fruit that were severely affected by the disease imparted a negative flavor to the juice. When fruit with normal appearance, harvested from HLB-affected trees were juiced, the juice flavor was not different from juice made from fruit harvested from healthy trees. It is important to the citrus processors to now know that they only need to sort out symptomatic fruit (small, green and lopsided fruit) from their processing line to maintain juice quality. Studies are now ongoing to determine threshold levels of HLB-induced off-flavors in normal juice. (NP 306, C 1, P.S. 1c, P.M. 2.1.2)

Exposure of simulated supermarket continuous light during simulated retail marketing of spinach increases vitamin concentrations. Human health benefits derived from consuming fruit and vegetables are due to the many human bioactive compounds (vitamins C, folate, E, K, and pro-vitamin A) found in produce. However, concentrations of these bioactive compounds are heavily influenced by plant genetics and pre/postharvest environmental display shelf-life. Scientists at Weslaco, TX, used spinach leaves from two popular commercially grown cultivars (Lazio and Samish) and placed them in plastic containers that were stored under continuous light or dark conditions. All vitamins increased in concentration with storage under continuous light and decreased under continuous dark. This research established, for the first time that, at least with spinach, the concentrations of human bioactive compounds in postharvest green (photosynthetic) tissues exposed to supermarket light levels (retail display) will increase during storage. This information should deter some of the concerns about diminishing nutritional quality of produce in retail settings. (NP 306, C 1, P.S. 1c, P.M. 2.1.2)

<u>Grass genome sequenced.</u> ARS scientists, in collaboration with Department of Energy (DOE) and other researchers, completed the sequencing and annotation of the entire *Brachypodium* genome. A paper describing the results was published in *Nature* and the genomic information is now publically available on several databases. In addition, a project to resequence additional accessions was initiated. To date, four lines have been resequenced and the analysis of the sequences has been initiated. Knowledge of the genome sequence of *Brachypodium* and the linear order of genes in the genome relative to other grasses will help researchers improve traits in energy crops and grain species. (NP 213, C1, P.S. 1b; P.M. 2.1.1; #5325-21000-017-00D)

New process for producing hydrocarbon fuels from biomass. A commercially-viable process for converting cellulosic biomass into drop-in replacements for petroleum-derived fuels would be a game-changing advancement for the biofuels industry. ARS scientists have combined the fermentative production of volatile fatty acids (VFAs) from biomass by ruminal bacteria with subsequent electrolysis of the VFAs to produce liquid hydrocarbons and hydrogen gas. The fermentation can be performed on ground biomass without additional pretreatment and without sterilization of the biomass or the culture medium. The electrolysis can be conducted at low voltages with inexpensive graphite electrodes. A U.S. patent application has been filed. (NP 213, C 3a, P.S. 3a1 and 3a2; P.M. 2.1.1; #3655-41000-006-00D)

Biorefinery-derived stimulators for butanol fermentation. Butanol is an advanced biofuel more compatible with the Nation's transportation fuels infrastructure than ethanol. ARS scientists have discovered that furfural and hydroxymethylfurfural, which are produced when biomass is pretreated with dilute acids, stimulate the rate of butanol fermentation by a factor of two or more. This discovery lowers the cost of producing butanol from any plant-derived feedstock. (NP 213, C 3a, P.S. 3a1; P.M. 2.1.1; #3620-41000-149-00D)

New, highly-efficient hemicellulase enzyme. Beta-D-xylosidase from the bacterium *Selenomonas ruminantium* is the most efficient enzyme for releasing the sugar, xylose, from biomass, but high concentrations of xylose inhibit this enzyme. ARS scientists developed a mutated enzyme which tolerates 300% higher sugar concentrations and thereby lowers the production costs for cellulosic ethanol. (NP 213, C 3a, P.S. 3a1; P.M. 2.1.1; #3620-41000-133-00D)

Genetic tools for grass breeding. ARS scientists in collaboration with CSIRO (Australia) used a high-throughput phenotyping platform (phenomics) to characterize over 100 natural accessions of *Brachypodium*, a model grass. Extensive natural variation in several traits relevant to biofuels including cell wall composition, stem density and fermentability have been found. ARS researchers also created over 4,000 T-DNA lines this year and released over 4,000 T-DNA lines made in the prior year to the public through a newly established T-DNA website. These tools will identify genes in bioenergy crops which can be manipulated to improve biofuel production. (NP 213, C 1, P.S. 1b; P.M. 2.1.1; #5325-21000-017-00D)

<u>Pyrolysis economics</u>. Unlike other biomass conversion technologies, pyrolysis may enable biorefining processes on or near the farm at a relatively small scale, thereby minimizing the costs associated with transporting large quantities of low-density biomass. By developing and using an ASPEN+ process cost & simulation model for biomass fast-pyrolysis, ARS researchers found that a 200 tons-per-day plant is the smallest size that would be competitive with \$85/bbl petroleum. (NP 213, C 3b and 3d, P.S. 3b2, 3b3 and 3d1; P.M. 2.1.1; #1935-41000-082-00D and #1935-41000-083-00D)

<u>Increasing yield of ethanol from corn stover</u>. One reason why cellulosic ethanol is much more expensive than corn-based ethanol is that biomass contains both hexoses, such as glucose, and pentoses, such as xylose. Corn-based ethanol is produced with brewers yeast, which converts only glucose. Although new

microorganisms have been developed to convert both hexoses and pentoses to ethanol, these recombinant organisms ferment glucose preferentially and do not begin to metabolize pentoses until low glucose concentrations have been reached. As a result, fermentation times are long and the pentoses are not fully converted. To overcome these hurdles, researchers at ARS and Iowa State University co-developed a two-stage simultaneous saccharification and fermentation process. In the first stage, pentoses are released and fermented to ethanol using an organism capable of highly efficient pentose metabolism; in addition, glucose is released and simultaneously converted to ethanol with brewers yeast. Using this process, an ethanol yield of 85 gallons per ton was achieved from corn stover. If the traditional process that ferments only glucose had been used, the yield would have been only 65 gallons per ton.

(NP 213, C3a, P.S. 3a1; P.M. 2.1.1; #1935-41000-083-00D)

<u>Distillers dried grains with solubles (DDGS) as fish feed</u>. Fishmeal, the traditional feed for aquaculture, has become very expensive (~\$1,500/ton) due to declining marine stocks. ARS researchers, in collaboration with South Dakota State University, determined that a feed combination of 40% DDGS (a coproduct of corn ethanol production), 9.5% soybean meal and 24% fishmeal resulted in the highest weight gain in Yellow Perch. Replacing the current diet in Yellow Perch aquaculture (40% fishmeal) with the DDGS-based diet could save the industry nearly \$9 million annually. (NP 213, C 3a, P.S. 3a4; P.M. 2.1.1; #5447-41000-003-00D)

<u>Phosphate-rich biochar</u>. Although poultry manure is rich in phosphorous, data were not available on whether charred manure would reduce the bioavailability of its phosphorus in soils. ARS scientists showed that poultry litter chars used as soil amendments in fact released a significant portion of their phosphorous. This finding shows that char can be used to sequester carbon while also acting as a source of phosphorous to plants. (NP 213, C 2 and 3b, P.S. 2c and 3b4; P.M. 2.1.1; #6435-41000-089-00D)

Distillers dried grains with solubles (DDGS) as a high-protein, high-fiber food additive. ARS researchers, in collaboration with South Dakota State University, conducted studies using various blends of DDGS (a coproduct of ethanol production) in Asian flat breads (naan and barbari). DDGS, which cost only \$0.05/lb, were found to replace corn-based food ingredients, which cost ~\$3/lb, at levels up to 20% with only minimal reductions in bread performance and consumer acceptability. Successful commercial use of DDGS as a human food ingredient represents an opportunity to increase the value of DDGS by nearly 60 times. (NP 213, C 3a, P.S. 3a4; P.M. 2.1.1; #5447-41000-003-00D)

Switchgrass grown for biomass energy results in significant soil carbon sequestration. The impact of growing large acreages of switchgrass on soil carbon is important in assessing the environmental consequences of biofuels production. In 1998, ARS scientists at Lincoln, Nebraska and Ft. Collins, Colorado initiated a switchgrass soil carbon sequestration study in eastern Nebraska. The study included two switchgrass cultivars, three nitrogen fertilizer rates, and two harvest treatments. In the nine years following grass establishment, soil carbon increased at rate of 0.9 U.S. tons per acre per year in plots in which best management practices were used. Biomass yields and carbon sequestration was significantly greater in plots in which nitrogen fertilizer was used than in plots where no fertilizer was applied. These results build upon switchgrass soil carbon sequestration data previously obtained in a five year study on 10 farms in Nebraska, South Dakota, and North Dakota. These finding demonstrate that growing large acreages of switchgrass will sequester significant quantities of carbon while producing high quality biofuel feedstocks. (NP213 C2 PS-A; P.M. 6.3.1, #5440-21000-028-00D)

<u>Corn stover for bioenergy</u>. Corn stover is the single most abundant crop residue biomass resource, and increasing ethanol yield from stover would improve the profitability of cellulosic ethanol systems. Relationships among grain yield and cellulosic ethanol traits were examined by ARS and University of Minnesota scientists at St. Paul, MN, and genetic markers for cell wall traits important for cellulosic ethanol production were identified. All the cellulosic ethanol traits (cellulose, lignin, and glucose release)

had moderate to high heritability and were not negatively related with grain yield, and genetic markers were found for all cell wall traits. The results indicate that it should be possible to breed corn for improved stover cellulosic ethanol traits while continuing to select for increased grain yield, and that marker-assisted selection could be used to speed corn stover improvement. Simultaneous improvements in corn yield and stover quality will allow significant increases in overall ethanol yield and net energy efficiency of biofuel production using corn. (NP213 C1 PS-B; P.M. 2.1.1; #3640-12210-001-00D)

Livestock Production (ARS Goal 2) (101, 106)

Select Examples of Recent Progress:

Turkey genome assembly completed for the industry. Scientists from ARS, Virginia Tech's Bioinformatics Institute and the University of Maryland's Center for Bioinformatics and Computational Biology completed the genomic map of turkey which will serve as the cornerstone for the development of sophisticated genomic technologies for the turkey industry. The turkey genome assembly was further strengthened when physical, comparative, and genetic maps built by researchers from Michigan State University and the University of Minnesota were used to match the DNA sequences to the turkey chromosomes. At completion, the original partnership expanded to include 68 scientists affiliated with 28 national and international research institutions. This project illustrates the rapid pace of genomic sequencing technology improvements. The turkey genome was sequenced in less than a year at a fraction of the cost of the chicken or bovine genomes. The information gleaned from this project will help breeders develop improved commercial turkey lines and significantly increase genetic progress for economically important traits, increasing the value of turkey products for both producers and consumers. (NP101, C 1, P.S 1C, P.M. 1.2.3, Project # 1265-31000-093-00D)

Commercial genotyping tools impact bovine livestock research and the dairy artificial insemination industry. A Single Nucleotide Polymorphism (SNP) is a variation or mutation in a DNA sequence occurring when a single nucleotide in the genome differs between members of a species or paired chromosomes in an individual. SNPs are responsible for much of the genetic variation between and within a species. ARS led in the development of a low density (3000 SNPs) and a high density (777,000 SNPs) beadchip product. Their impact on the industry has been dramatic and will become more so as the cost continues to decrease for the SNP beadchips. The BovineSNP50 assay, (a beadchip with 50,000 SNPs), developed by ARS, remains the global standard for cattle genomics research and genetic prediction use, with sales surpassing 500,000 samples. ARS received the USDA Secretary's Honor Award for Excellence in 2010 for implementing genome selection in dairy cattle using these technologies. Adoption of these technologies has had a tremendous economic impact on the dairy industry and has resulted in significantly improved genetic prediction accuracies and enhanced genetic progress. (NP101, C 1, P.S 1A, P.M. 1.2.3, Project # 1265-31000-098-00D)

Triple-acting staphylolytic peptidoglycan hydrolase fusion proteins developed as potential alternatives to conventional antimicrobials. Staphylococcus aureus is a notorious pathogen that has a high rate of antimicrobial resistance development. However, it is believed that no bacterium can resist three simultaneous, uniquely-acting, antimicrobial agents. To develop such antimicrobials ARS fused the staphylococcal phage K endolysin LysK, to the staphylolytic bacteriocin, lysostaphin. These two proteins together harbor three unique, synergistic peptidoglycan hydrolase enzymatic domains. The resultant triple-acting fusion protein maintains all three enzymatic activities in the final fusion. When Staphylococcus aureus cultures were exposed repeatedly (>10X) to sublethal doses of this triple fusion, no resistance was observed, despite significant resistance observed in cultures exposed to either LysK or lysostaphin alone. Staphylococcus aureus naturally resides topically on humans and animals including

the nasal nares. The triple-acting staphylolytic peptidoglycan hydrolase fusions perform with greater efficacy than either of the parental lysins in eradicating Staphylococcus aureus in a rat model of nasal colonization. We predict these triple fusion antimicrobials may potentially result in the development of a new class of antimicrobials for the livestock industries which can be used to treat disease and increase production efficiencies without creating antimicrobial resistance. (NP101, C 1, P.S 1A, P.M. 1.2.2, Project # 1265-31000-091-00D)

Copy number variation (CNV) and segmental duplication regions of the bovine genome. A copy number variant (CNV) is a segment of DNA in which differences in base pairs or even genes exist between two or more genomes. The segment may range from one kilobase to several megabases in size. Animals ordinarily have two copies of each autosomal region, one per chromosome. CNVs may either be inherited or caused by mutation. CNVs can be caused by genomic rearrangements such as deletions, duplications, inversions, and translocations and are a surprising and important source of genetic variation not associated with traditional genetic theory. ARS research has identified over 200 candidate CNV regions (CNVRs) in total and 177 within known chromosomes, which harbor, or are adjacent to, gains or losses in genetic material. These 177 high-confidence CNVRs cover ~1.07% of the genome. Multiple gene families have been identified and this research detected and confirmed marked differences in CNV frequencies across diverse breeds, indicating that some cattle CNVs are likely to arise independently in breeds and contribute significantly to breed differences. Regions with varying copy number span about 400 known genes for specific biological functions, such as immunity, lactation, reproduction, and rumination. These results provide a valuable resource beyond microsatellites and single nucleotide polymorphisms to explore the full dimension of genetic variability for future cattle genomic research and will lead to significant genetic tools for the beef industry. These tools will increase genetic progress for traits of economic value, illuminate variability in phenotypic trait expression, explain complex production and behavioral traits, and improve disease resistance and treatment strategies. (NP101, C 1, P.S 1A, P.M. 1.2.3, Project # 1265-31000-098-00D)

Infrared beak trimming improves hen welfare. Beak trimming, using a hot blade, is a common practice in the poultry industry that is often criticized as potentially painful or inhumane. ARS researchers investigated an alternative method of beak trimming which uses an infrared laser similar to those used in biomedical procedures. In this research both laser and hot blade trimming treatments were applied on production hens from 5 to 35 weeks of age. Hens which were beak-trimmed using the infrared laser method showed an improvement in performance and a reduction in stress levels. These results suggest that the infrared beak treatment provides a more welfare friendly means of beak trimming, allowing birds to display more efficient feeding behavior with less morphological abnormalities of the beak stumps. This study provides scientific evidence to support the use of infrared beak-trimming by producers and addresses the welfare concern of the current practice for the poultry industry. (NP101, C 2, P.S 2A, P.M. 1.2.1, Project # 3602-32000-009-00D)

Microbial diversity in the bovine rumen. Scientists at ARS and the J Craig Venter Institute completed a survey of the unique diversity of microbial populations in the rumen of cows fed forage diets and established protocols for use in future investigation. This work established guidelines for conducting future research examining the rumen micro-biome. The observed bacterial diversity was between 3181 to 7483 species-level taxonic units (individual organisms), the largest number observed for a ruminant species. This diversity was dominated by Bacteroidales, unknown bacteria, Clostridiales and Bacteroidetes. There was a considerable difference in microbial diversity between cows from which the samples came. The fungal community was sequenced to saturation and also resulted in identification of a novel unknown fungal group. Raw extrusa should be suitable for future studies on rumen microbial diversity. These results will help researchers as they seek to improve the efficiency of dairy cattle diets and minimize the output of greenhouse gases by dairy cattle through manipulation of the bovine ruminal

community. (NP101, C 2, P.S 2C, P.M. 1.2.3, Project # 5434-31000-016-00D)

Marek's disease virus evolves to higher virulence in poultry with limited genetic variation. Marek's disease (MD), a serious problem for the poultry industry caused by the pathogenic Marek's disease virus (MDV), is primarily controlled by vaccines. However, MD is now increasing in impact as the MDV continues to evolve to higher virulence. Most studies addressing the evolution of MDV virulence have concentrated on the virus while largely ignoring the hosts' influence. The host system, called the major histocompatibility complex (MHC), represents a highly variable system designed to defend a species from extinction from the fast paced evolution of a parasite. In natural chicken populations, there are hundreds of different MHC haplotypes that oscillate or rotate in response to pathogen evolution providing protection and resistance. A haplotype is a specific and preserved combination of alleles (DNA sequences) at different places (loci) on the chromosome that are transmitted together. A haplotype may be one locus, several loci, or an entire chromosome and in this case represents the MHC. Commercial poultry breeding has limited the number of MHC haplotypes to six or less which restricts the ability of the poultry line to adapt to the disease challenge -i.e., the proper MHC haplotype does not exist. ARS research shows that MDV can evolve to higher virulence in birds with a single MHC haplotype. Based on this research ARS recommends that producers rotate the placement of MHC haplotypes by rotating genetic lines, similar to the simple method of crop rotation used to control pests in the field. This simple production practice should prevent the increase in virulence in MDV and to reduce the chronic problem of MD for producers resulting in substantial savings to the poultry industry and increased performance and production efficiencies. (NP101, C 1, P.S 1B, P.M. 1.2.3, Project # 3635-31320-008-00D)

Rumen-protected methionine allows cows to produce more milk on less protein. Dietary protein supplies lactating cows with amino acids to make the protein in milk and body tissues. About half of the amino acids are essential, meaning they cannot be made by the cow but must be supplemented and are absorbed in the intestine. Methionine, often the most limiting essential amino acid, is available in rumen-protected form allowing absorption at the intestine. A new, cheaper type of protected methionine has just become commercially available. This compound is a chemically modified derivative of methionine that can resist microbial attack in the rumen and is converted to methionine in the cow's body after absorption. Lactating dairy cows fed a 15.6% protein diet were given no methionine, methionine protected with a physical coating, or the new chemically protected form of methionine. None of the diets influenced feed intake or actual milk production. However, feeding the chemically protected form of methionine increased yield of energy-corrected milk and milk protein concentration. Moreover, cows tended to yield more fat and protein on either methionine source versus low or high protein diets without methionine. Feeding 16.8% protein without methionine elevated urinary nitrogen excretion and reduced nitrogen efficiency from nearly 35% to about 30%. Results with either methionine source were similar, indicating that the less expensive chemically protected form was as effective as the physically protected methionine. This research indicates that protein efficiency is increased and the potential for nitrogen pollution on U.S. dairy farms is reduced by an inexpensive supplement of rumen-protected methionine. (NP101, C 2, P.S 2C, P.M. 1.2.2, Project # 3655-31000-021-00D)

A method for determining whether piglets receive adequate colostrum from the sow. Preweaning mortality of piglets represents a substantial loss to swine producers. One possible factor contributing to this loss is the failure of neonatal piglets to obtain sufficient colostrum from the sow which can be caused by either failure of the piglet to nurse, or failure of the sow to initiate lactation. An inexpensive and rapid method, the "immunocrit," was developed and validated by ARS scientists to measure newborn piglet serum immunoglobulin G (IgG), which reflects whether a piglet has received adequate colostrum. Results indicated that immunocrit measurements are predictive of piglet mortality, and litter average immunocrit indicated the ability of the sow to transmit IgG (via colostrum production). Low immunocrit values were primarily due to the failure of individual piglets to nurse and not due to failure of the sow to produce colostrum. Litter average immunocrit can be used to identify sows that fail to initiate colostrum

production enabling selection for efficient IgG transfer (presumably efficient colostrum production) from sow to piglet. The immunocrit has been adopted by industry to monitor and manage newborn piglet colostrum intake which will increase piglet survivability to weaning, increasing production efficiencies for the pork industry. (NP101, C 2, P.S 2B, P.M. 1.2.2, Project # 5438-31000-084-00D)

Identification of DNA markers associated with respiratory disease in cattle. Previously, ARS scientists had established that bovine chromosome 20 is the site of genetic variation affecting a range of infectious disease-related phenotypes, using new high-density genotyping arrays. Now they have examined the annotated genome of cattle in the area of this variation and identified two functional candidate genes with known roles in immune function, with gene symbols ANKRA2 and RP105. Previously discovered DNA sequence markers not present in the initial arrays, but predicted to lie in this area of the genome, were tested for association with disease traits in order to provide supporting data for the original study, and identify markers with better predictive merit for disease resistance. Seven markers in the two genes displayed association, providing potential markers for selecting animals with superior response to respiratory disease challenge in beef cattle. These technologies promise to lower animal mortality and morbidity for beef producers, increasing production efficiencies and improving animal well-being. (NP101, C 1, P.S 1B, P.M. 1.2.3, Project # 5438-31000-085-00D)

Noninvasive tenderness prediction system accurately predicts tenderness of most major beef muscles. The U.S. beef industry and the Agricultural Marketing Service (AMS) have sought implementation of standards for tenderness. ARS scientists previously developed a noninvasive method to predict tenderness of the ribeye muscle of beef carcasses based on visible and near infrared (VISNIR) spectroscopy. Under a research agreement between ARS and the National Cattlemen's Beef Association, ARS scientists determined that the existing system could also predict tenderness of other muscles of the carcass. Similar results were obtained with the application of VISNIR directly to the exposed *gluteus medius* on the anterior end of top sirloin subprimals, either during carcass fabrication or after aging. These results suggest this technology can be efficiently and cost effectively utilized by industry to control variation in tenderness which will greatly enhance consumer acceptance and consumption of U.S. beef products. (NP101, C 3, P.S 3A, P.M. 1.2.2, Project # 5438-31430-004-00D)

National Animal Germplasm Program (NAGP) – FY2010 Collection Summary. The national collection of animal genetic resources, held by ARS, ended FY10 with 599,112 germplasm and tissue samples from 13,638 head of agronomically important animal species. Over the past five years the collection has grown linearly and increased in size by 163%. To date, the collection contains samples from 32 different species, 143 breeds, and 181 specialized lines across all life forms in the collection. The collection is the largest national repository in the world and is at least twice as large as any of the European gene bank collections. This repository serves a vital role in protecting and preserving economically valuable germplasm and genetic diversity for the livestock industries in the U.S. and around the world. (NP101, C 1, P.S 1C, P.M. 1.2.4, Project # 5402-31000-004-00D)

Improved catfish feed conversion through pond oxygen management. Dissolved oxygen (DO) is the most critical water quality parameter in warmwater aquaculture. Controlled studies on the impact of DO fluctuations on channel catfish have been lacking. ARS researchers at Stoneville, Mississippi examined the impact of DO concentrations on catfish growth, food consumption, and food conversion. Results showed that higher DO concentrations (2.5-3.0 milligrams per liter) are required for optimum food conversion and growth, and this improved growth will significantly shorten the production cycle. Increased growth resulting from improved DO management can reduce food conversion ratios from an estimated industry-wide 2.5-3.0 down to 2.0, greatly improving the profitability of catfish farming. (NP 106, C 2, P.S. 2C, P.M. 2.2.2, Project #6402-13320-004-00D)

<u>Arctic charr selected for increased growth</u>. Arctic charr have a flavor many consumers feel is superior to trout and salmon. Scientists at the National Cold Water Marine Aquaculture Center in Franklin, Maine

evaluated the growth of two different arctic charr stocks for culture in recirculating aquaculture systems. Development of a breeding program for U.S. arctic charr stocks could alleviate some of the production problems limiting expansion of arctic charr culture and provide a source of germplasm with improved production traits. A charr line selected for fast growth and delayed sexual maturity will be released for cooperative research evaluations and commercial production in the fall of 2010. (NP 106, C 1, P.S. 1C, P.M. 2.2.2, Project #1915-31000-003-00D)

Split-pond aquaculture systems may increase catfish production by two to three times over traditional earthen ponds. A split-pond aquaculture system has been developed that may increase channel catfish production by two or three times that achieved in traditional earthen ponds. The new system splits an existing earthen pond into two unequal sections with an earthen levee and then links the two systems by circulating water that is pumped with a large, efficient, slow turning paddlewheel. Fish are held in the small section; the larger section provides waste treatment and oxygen production. Annual catfish production has averaged more than 15,000 kilograms per hectare during the study. Several commercial growers have implemented similar systems. (NP 106, C 5, P.S. 5B, P.M. 2.2.2, Project #6402-31630-001-00D)

<u>Improved soy concentrate for aquafeeds</u>. Soy protein concentrate (SPC) is a highly nutritious ingredient in aquafeeds, but is currently too expensive to be practical. ARS researchers in Aberdeen, Idaho, have developed a modified method for the production of feed grade SPC and are working with a commercial research partner in pilot scale testing and possible commercialization. Availability of a feed grade SPC will make fish meal free diets more cost effective, and the aquaculture industry more sustainable. (NP 106, C 1, P.S. 1A, P.M. 2.2.2, Project #5366-21310-004-00D)

Development of modified live Edwardsiella tarda and Aeromonas hydrophila vaccines for prevention of diseases in aquaculture. ARS researchers at Auburn developed and patented Edwardsiella tarda and Aeromonas hydrophila_vaccines under a Cooperative Research and Development Agreement (CRADA) with a vaccine manufacturer. These modified live vaccines can be effectively used by a bath immersion method to cost effectively immunize large numbers of fish. The new A. hydrophila vaccine may be especially useful in preventing huge losses of food size catfish (over one million pounds in 2009) that caused considerable economic hardship to Alabama catfish producers. (NP 106, C 2, P.S. 2G, P.M. 4.2.2, Project #6420-32000-019-00D)

Year-round spawning achieved with pompano. Lack of sustained year-round production of juveniles for grow-out operations is one of the foremost bottlenecks of marine finfish aquaculture. Spawning induction protocols were developed and tested. Spawning performance of Florida pompano broodstock, measured as number of eggs, fertilization, egg quality, and hatch rate was quantified over a 12 month period. Spawning was achieved in 10 months of the year having an average production of 1.9 million eggs/year with no discernable diminishment in egg quality over time. This work demonstrates Florida pompano seedstock can be produced year-round from a small population of broodstock, and overcoming one of the key bottlenecks to marine finfish aquaculture. (NP 106, C 2, P.S. 2B, P.M. 2.2.2, Project #6225-63000-008-00D)

Laboratory infection models to assess pathogenesis of Viral Hemorrhagic Septicemia. Viral Hemorrhagic Septicemia (VHS) emerged as a new disease threat to fish in the Midwest in 2003. Lack of tools and infection models has hampered development of countermeasures against this disease. Over the past two years, collaborations among scientists from ARS; University of Wisconsin-Milwaukee; and the U.S. Geological Survey (USGS), Western Fish Research Center, Fish Health Laboratory have led to development of the standard model for the Midwest VHS virus strain using the yellow perch. Standardized exposure route, viral dose range, and exposure time for subsequent studies to evaluate differences in yellow perch broodstock VHS susceptibility have been determined. These methods will

enable systematic testing of vaccines and development of countermeasures. (NP 106, C 2, P.S. 2D, P.M. 4.2.1, Project #3655-31320-001-00D)

Developing non-traditional fish feeds for water recirculating systems. The interest and usage of alternative protein sources for fish feed is increasing as the aquaculture industry expands beyond the sustainable limits of marine protein sources used in traditional diets (fishmeal). Experimental plant protein based diets were tested on rainbow trout by ARS scientists at Leetown and Shepherdstown, West Virginia, to assess fish growth rate, feed conversion, final size, and survival, as well as water quality, relative to fish fed traditional diets. No significant differences were found between the two groups. All water quality parameters tested were within safe limits, however higher concentrations of total suspended solids, carbonaceous biochemical oxygen demand, and total nitrogen and nitrate nitrogen were measured within systems fed the plant protein-based diet. These findings indicate significant progress toward a more sustainable non-fishmeal-based diet as reflected by the comparable trout performance on the two diets; findings also suggest that the recirculating flow should be treated with ozone to reduce the suspended solids and organic carbon accumulation that can occur when using plant-protein-based feed. (NP 106, C 2, P.S. 2A, P.M. 2.2.2, Project #1930-31320-001-00D)

Molecular tests for stress tolerance genes in Pacific oysters. Selective breeding of Pacific oysters to improve agronomic performance is in early stages. Scientists from the ARS Shellfish Genetics Program in Newport, Oregon demonstrated that the expression levels of stress-related genes in juvenile Pacific oysters are predictive of subsequent growth and survival in field trials. This research indicates that molecular testing in the laboratory may provide a rapid, low-cost method for identifying superior performance in the field. These assays will help scientists predict performance and accelerate genetic improvement efforts. (NP 106, C 1, P.S. 1D, P.M. 2.2.2, Project #5358-31000-002-00D)

Determination of nutritional requirements for Pacific threadfin (moi). Pacific threadfin, known in Hawaii as moi, is a fish with considerable aquaculture interest; however the nutrient requirements are not well defined. Protein is the most expensive ingredient in feed. Therefore it is important to optimize the protein and energy levels in feed, and meet the requirements for all essential amino acids in order to reduce feed costs. This will also minimize use of dietary protein for energy and decrease ammonia discharge that impacts the environment. Scientists at the Oceanic Institute conducted feeding studies that showed that diets containing 35% protein and 14% lipid generated the best growth performance. In a tandem study, the requirement for lysine, a critical and often the first limiting amino acid in ingredients used for fish feed, was determined. The results provide basic information to estimate nutritional requirements, which are essential for developing cost effective diets for moi culture. (NP 106, C 3, P.S. 3A, P.M. 2.2.2, Project #5320-31000-008-00D)

Crop Production (ARS Goal 2) (301, 302, 305)

Select Examples of Recent Progress:

NP301

<u>Cacao genome sequenced</u>. Cacao, the source of chocolate, is a multi-billion dollar international commodity grown by several million small farmers in tropical developing nations. Threatened by many virulent diseases and damaging pests, cacao requires new tree types with inherent resistance to pests and diseases, plus high yields and fine cocoa quality. Currently, cacao breeders lack DNA genetic markers required for rapid selection of trees with desired traits at the seedling stage, rather than at maturity. ARS scientists in Miami, Florida and Stoneville, Mississippi, with collaborators at Mars, Inc., IBM, and several U.S. universities enlisted a novel mixture of traditional and leading edge techniques to fully sequence the

genome of a particular cacao variety that shares ancestry with many of the trees grown worldwide. This genome sequence can now be compared with genetic information from other cacao varieties with different properties to rapidly identify many thousands of genetic markers, and thereby accelerate cacao genetic improvement to benefit farmers and cocoa processors globally.

(NP301; C2, PS 2B; PM 2.2.3; Project # 6631-21000-017-00D)

New sorghum germplasm developed with improved value as a bioenergy feedstock. Sorghum use as a bioenergy feedstock can be improved by increasing biomass digestibility. ARS scientists in Lincoln, Nebraska have determined the effects of inserting and combining brown midrib mutations into a grain sorghum hybrid. These mutations lowered the content of lignin, a polymer that provides rigidity to the cell wall and hinders cell wall decomposition, in a grain sorghum hybrid. Effects of the genetic changes to sorghum biomass composition and whole plant physiology were assessed in a two year field experiment. Overall, the results demonstrated that sorghum brown midrib mutants have reduced lignin and increased biomass digestibility while having a minor impact on plant fitness and yield in hybrid backgrounds. This sorghum germplasm with reduced lignin content and increased biomass digestibility provides a new genetic resource to develop sorghum for use as a bioenergy feedstock. (NP301, C3, PS 3C; PM 2.2.3; Project # 5440-21220-027-00D)

HoneySweet plum conditionally registered by EPA. There are limited sources of Plum Pox Virus (PPV) resistance in stonefruits. "HoneySweet," a plum genetically engineered for resistance to PPV, which had previously been deregulated by the Animal and Plant Health Inspection Service (APHIS) and the FDA, is now conditionally registered by EPA. HoneySweet is the result of over 20 years of research by ARS researchers in Kearneysville, West Virginia and other ARS and European collaborators. It has been found to be resistant to PPV in test plots in Europe over the last 10 or more years. When commercially available, it will be the first genetically engineered disease resistant temperate fruit tree available to U.S. growers, providing a high quality, fresh market, PPV-resistant plum. It can also serve as a breeding parent to reliably and efficiently introduce the resistance trait into new plum varieties. (NP301; C3, PS 3C; PM 2.2.3; Project # 1931-21000-017-00D)

Development of "intragenic" potatoes containing a late blight-resistance gene. The commercial introduction of crop plants improved by using biotechnology has been limited by problems associated with public perceptions of transgenic foods. These limitations are being addressed in part by development of novel methods for in vitro genetic modification, referred to as "intragenic" technology. This method of gene introduction results in all native transgenic lines, i.e., lines that contain no foreign DNA. ARS scientists in Albany, California, in cooperation with scientists at the J.R. Simplot Co., in Boise, Idaho have successfully developed intragenic potatoes that contain a gene from wild potatoes known to confer resistance to the most devastating of potato diseases, late blight, the cause of the Irish potato famine which still plagues growers today with new, virulent genotypes. These potatoes do not contain any non-potato DNA, and were generated without using selection (i.e., they do not contain antibiotic/herbicide resistance genes as markers). Intragenic lines with late blight-resistance in the greenhouse and under field conditions will be an important option for preventing losses to late blight. (NP301; C2, PS 2C; PM 2.2.3; Project # 5325-21420-004-00D)

New germplasm and genetic resources developed to protect wheat and barley from the Ug99 stem rust. Wheat and barley germplasm with resistance to the virulent Ug99 stem rust strain is urgently needed to protect the global grain supply. ARS researchers in Raleigh, North Carolina, Aberdeen, Idaho, and St. Paul, Minnesota, organized and evaluated 4,000 wheat and barley varieties and germplasm lines in Njoro, Kenya, for resistance to Ug99 stem rust. Lines were submitted from more than 25 public and private sector U.S. breeding programs. ARS researchers in Raleigh also developed 750 wheat lines having stem rust genes combined in two, three, four, and five gene stacks (or pyramids). These lines also have gene combinations for leaf and stripe (yellow) rusts. The researchers in Raleigh also developed and distributed

30 advanced lines of wheat having multiple-gene resistance to stem rust race Ug99 to wheat breeders in 32 countries in cooperation with the International Wheat and Maize Improvement Center in Mexico. This information will enable breeders in the United States to identify and deploy resistance to Ug99 stem rust in advance of the pathogen ever arriving in the United States. These lines will greatly aid U.S. and international wheat breeders develop better worldwide resistance to stem rust race Ug99. (NP301, C3, PS 3B & 3C; Project # 6645-22000-016-00D)

New approaches to citrus improvement through "intragenics". The method of genetically transforming crops using genes derived from that particular crop, termed "intragenics", has been targeted as a commercially viable means of developing resistance to the devastating Huanglongbing (HLB) or citrus greening disease. But first this requires the development of a citrus genome database for its application. The limited amount of publicly accessible citrus genomic DNA sequence has significantly impeded application of this process to citrus improvement. To support the utilization of this technology to address critical issues in citrus production, ARS scientists in Albany, California, and Fort Pierce, Florida, partnered to sequence the genome of the citrus rootstock 'Carrizo', and made this sequence publically available (http://citrus.pw.usda.gov/). While this database will be useful in a broad array of applications, ARS researchers are employing it to identify citrus DNA sequences useful in expressing HLB-resistant genes. For example, this data has been employed to identify DNA sequences to allow high-level expression of introduced genes in vascular tissue, where the causal agent of HLB is found. Given that HLB currently represents the most serious threat to citrus production in the United States and no resistance genes have yet been identified, the successful development of resistant trees offers a potential solution crucial in sustaining this crop. (NP301; C2, PS 2C; PM 2.2.3; Project # 5325-21420-004-00D)

Bean development for use under high temperatures. Common bean (*Phaseolus vulgaris L.*) is a vital part of the diet in many areas of the world. Bean reproductive development is particularly sensitive to high temperature stress, resulting in yield reduction and limited adaptation to warmer climates. Two kidney beans were developed by ARS scientists in Mayaguez, Puerto Rico, with cooperators at the University of Puerto Rico, Cornell University, and the University of Tennessee, that are tolerant to high temperature conditions. One variety is tolerant to high day and night temperature stress, while the second is tolerant to high daytime temperature stress and moderate nighttime temperature stress. The use of these kidney beans can improve yield under hot summer conditions for farmers in regions prone to high temperature stress. In addition, they can be used for improving heat tolerance in other large-seeded beans through breeding and selection.(NP301; C3, PS 3B; PM 2.2.3; Project # 6635-21000-048-00D)

The new soybean genome sequence is linked to 30 years of trait information at SoyBase and the Soybean Breeders Toolbox. The soybean genome DNA sequence, produced by ARS scientists in Ames, Iowa, and Beltsville, Maryland, together with university and Department of Energy collaborators, was released in 2010 and is one of the largest and most complex genomes decoded to date. To take full advantage of this important new resource for soybean improvement, ARS scientists incorporated the sequence information into SoyBase and the Soybean Breeders Toolbox, the USDA's soybean genetics and genomics database. SoyBase now includes extensive 'under-the-hood' links to 30 years of data for many high-value traits, including seed yield, seed quality and disease susceptibility, biochemical pathways, and tens of thousands of new DNA marker sequences and genes, as well as powerful, new visualization tools for the soybean genetic and physical maps and the soybean genome sequence. SoyBase now will enable plant breeders to comprehend the genetic control and function of genes that govern critical agronomic traits. (NP301; C2, PS 2A, 2B & 2C; PM 2.2.3; Projects # 3625-21000-052-00D, 1275-21000-263-00D, and 3625-21220-004-00D)

<u>Discovery of 33,065 new simple sequence repeat DNA markers to enrich the soybean genome.</u> With the recent release of the whole DNA sequence of the soybean genome, the task at hand is now to identify large numbers of DNA markers across the 20 soybean chromosomes. ARS scientists in Beltsville,

Maryland, collaborating with ARS scientists in Ames, Iowa, screened the DNA sequence of the 20 soybean chromosomes and identified more than 33,000 simple sequence repeat (SSR) DNA markers with a high probability of effectiveness in discovering the positions of genes on the soybean chromosomes to enhance DNA marker-assisted soybean breeding. A database was created, which contains the information required for the use of these markers, including the specific position of each marker on the 20 soybean chromosomes. This information is publicly available on SoyBase (http://soybase.org), the USDA ARS Soybean Genome Database. The information will be useful in fine-mapping of genes, a process that involves identification of markers that are very tightly linked to a targeted gene. A genetic fine map of a specific gene locus will usually have a goal to identify and locate markers as close as possible on both sides of the targeted gene, and in soybeans, has been hindered by lack of the type of information presented by this discovery. Researchers can now acquire specific genes for further precise studies or for use in crop improvement by transgenic technology. (NP301; C2, PS 2B; PM 2.2.3; Project # 1275-21000-263-00D)

ARS scientists discovered natural variation that markedly increases Vitamin A in corn. Billions of people and livestock use corn (maize) as a major source of subsistence and nutrition. However, corn is generally not a good source of pro-Vitamin A, and vitamin A deficiency can lead to childhood blindness and immune deficiencies. Building on a prior discovery, ARS researchers in Ithaca, New York, and collaborators, discovered that the natural variation at two genes in corn can be exploited to breed corn with a 16-fold higher level of pro-Vitamin A than standard corn. Crop breeders are already starting to use this discovery to breed more nutritious varieties of corn, particularly where increased pro-Vitamin A is urgently needed. (NP301, C3, PS 3B; PM 2.2.3; Project # 1907-21000-029-00D)

New molecular breeding tools for heart-healthy oats. The oat genome is so large and complex that little genetic information and molecular markers have been available to assist oat breeding. This has meant that breeding for sustainable oat production and increased nutritional value has been slowed. In 2010, ARS researchers in Aberdeen, Idaho, and Albany, California, used innovative DNA sequencing methods to produce 650,000 oat gene DNA sequence fragments. In addition, ARS researchers in Aberdeen teamed with ARS scientists in Fargo, North Dakota, and Raleigh, North Carolina, to develop the first oat-based single nucleotide polymorphism (SNP) markers. More than 700 SNP markers, which are highly useful to plant breeders, have been developed. The research was supported by ARS with additional funding from General Mills, North American Millers Association, and the USDA National Institute of Food and Agriculture. These major advances in molecular markers are being made publicly available for all oat breeders. This plethora of new tools will enable oat breeders to use molecular markers for the first time to select and accelerate breeding for agronomic traits, disease resistance, and nutritional quality (high fiber and beta-glucans). (NP301; C2, PS 2C; PM 2.2.3; Project # 5366-21000-028-00D)

Compositionboards from cotton and guayule waste. Using biomass materials from cotton gin byproducts (CGB) and guayule wastes in value-added products can enhance the economic value of these crops, and help to alleviate waste management and environmental problems. ARS scientists in Lubbock, Texas, and Maricopa, Arizona, with a collaborator at the University of Illinois, evaluated important physical and mechanical properties of composition boards made from CGB and guayule waste. Although refinements are needed to further enhance the performance of these biomasses for specific composition board applications, these bio-based CGB and guayule waste boards showed great potential for use as construction materials. (NP301; C3. PS 3C; PM 2.2.3; Project # 5347-21410-005-00D)

<u>Release of new apple rootstocks</u>. ARS researchers in Geneva, New York, released four new apple rootstocks:

• G.214, a dwarfing, precocious, productive rootstock resistant to fire blight, with multiple disease resistance (fire blight, phytophthora root rot, and wooly apple aphid) that has performed very well

- in replant trials under organic management and is tailored for fresh market high density apple production amenable to mechanization.
- G.890, a semi-dwarfing productive plant resistant to fire blight that has performed well in difficult replant soils in Washington state and is tailored for fresh and processing apple production.
- G.210, is semi-dwarfing, has survived the series of inoculations with apple rootstock pathogens (*Erwinia amylovora*, *Phytopthora cactorum*, wooly apple aphid), and it has been characterized as tolerant to the replant disease complex in several replicated studies.
- G.969, a semi dwarfing productive plant, resistant to fire blight that lends itself as a good support for weaker or more difficult scion varieties like Honeycrisp.

All these rootstocks were tested in the field for productivity and precocity and performed very well when compared to other rootstocks that have similar vigor characteristics. All of these rootstocks have been transferred to the nursery industry for large scale production and are expected to have a major role in sustainable apple production for years to come. (NP301; C3, PS 3C; PM 2.2.3; Project # 1910-21000-023-00D)

Effects of citrus greening on citrus juice quality defined. When Huanglongbing (HLB) or citrus greening disease was confirmed in Florida, questions arose regarding impacts the disease may have on juice quality. There was belief that juice from diseased trees produced novel off-flavor compounds that would have negative effects on juice flavor. ARS researchers in Fort Pierce, Florida, compared chemical and flavor components of juice from healthy and HLB-affected trees, and determined that no novel off-flavor compounds were produced. Results indicate that the most consistent difference between juices from healthy or diseased trees was a reduction in total sugar content. Although trained taste panelists could distinguish between juice produced from healthy or diseased fruit, differences were minimal and related to less sweetness and more acidity. These results indicate that production of good-tasting citrus and citrus juice can continue with a high-quality product from existing cultivars even though they become infected with greening, as long as they are productive. This information is of critical importance to citrus grove managers and citrus juice processors in Florida where the majority of citrus production is for juice. . (NP301; C3, PS 3C; PM 2.2.3; Project # 6618-21000-013-00D)

New method developed to identify drought-tolerant sorghum lines. Sorghum yield losses to drought are significant, but crop breeding for drought tolerance under practical field conditions is difficult because of variable rainfall patterns. ARS scientists in Lubbock, Texas, have developed a break-through method to screen for drought tolerance in plants that are not even drought stressed. A new method, involving a short-term heat stress of leaf tissue followed by temperature recovery, is effective in identifying stay-green lines with drought tolerance. The method has proven effective in identifying more drought tolerant lines at both pre- and post-flowering and has been used to identify new sources of drought tolerance in a sorghum lines from Sudan. This new selection method will greatly reduce the selection time needed by crop breeders to develop more drought tolerant sorghum. (NP301; C2, PS 2C; PM 2.2.3; Project # 6208-21000-017-00D)

NP302

<u>Development of high oleic acid soybeans.</u> Soybean oil can be improved for its nutritional value in human foods and for industrial biodiesel use by altering its fatty acid composition. Vegetable oils with high oleic acid contents are desirable for the health benefits of the monounsaturated fatty acids which have recently made olive and canola oils very popular. High oleic acid content also dramatically improves oxidative and temperature stability of the oil, and improves cold flow properties in diesel engines. ARS scientists in Columbia, Missouri identified and combined mutant alleles of two soybean fatty acid desaturase genes resulting in high oleic acid soybean oil. The researchers developed a technology to directly select the

genes conferring the desired fatty acid profile, thus accelerating the rate at which new soybean varieties containing this important trait can be made available to producers. (NP302; C2, PS 2C; PM 2.2.3; Project # 3622-21000-029-00D)

New genetic information on corn kernel quality traits. Information on the genes and the genomic regions that control corn quality, including kernel starch, protein, and oil content, is needed by researchers to improve the nutritional and product quality of corn products. ARS researchers in Columbia, Missouri analyzed the kernel content of over 5,000 recombinant inbred lines from diverse types of corn. The lines were grown in seven locations. Researchers then used 1.6 million molecular markers and statistical analyses to identify regions of the genome that control each of the three kernel traits. The research resulted in successful identification of many genetic variants that strongly affect each kernel trait. Notably, new corn lines with a wide range in oil quality were identified. These results provide new tools and genetic resources to develop new corn lines and hybrids with a wide range of oil, starch, and protein content. (NP302; C1, PS 1B; PM 2.2.3; Project # 3622-21000-027-00D)

Discovery of natural variation in ozone tolerance in soybean. Ozone, the atmospheric pollutant, is responsible for billions of dollars in lost crop production each year. The effects of ground level ozone on soybean photosynthesis, seed yield, and antioxidant production, was investigated in 10 different soybean cultivars by ARS scientists in Urbana, Illinois. Doubling background ozone decreased soybean yields on average by 17 percent, but the variation in response among cultivars and years ranged from 8 to 37 percent. Chlorophyll content and photosynthetic parameters were positively correlated with seed yield, while antioxidant capacity was negatively correlated with photosynthesis and seed yield, suggesting a trade-off between antioxidant metabolism and carbon gain. Ozone exposure response curves indicated that there has not been a significant improvement in the tolerance of commercial soybean cultivars to ozone in the past 30 years. The discovery of genetic variation in ozone tolerance in non- commercial soybean cultivars is an important step toward improving ozone tolerance in commercial soybeans through breeding. (NP302; C2, PS 2B; PM 2.2.3; Project # 3611-21000-020-00D)

Discovery of the two genes responsible for the soybean low phytate trait. Much of the phosphate in soybean seeds is present as a component of phytic acid, an anti-nutritional factor which impedes the bio-availability of inorganic elements in food and feed containing soybean. This presents contamination issues via runoff from animal waste disposal. Determination of the molecular genetic basis of a soybean line containing the low phytate trait by ARS scientists in Columbia, Missouri resulted in the discovery of two mutant genes that lower phytate by acting in combination. An additional low phytate soybean line was also characterized and found to contain a dramatic mutation in one of the genes. A method was developed to directly select for the mutations discovered. Application of this method should simplify development and release of soybean varieties with improved nutritional quality as a trait important for feed and food applications. Such varieties would have a significant impact on the livestock protection industry. (NP302; C2, PS 2C; PM 2.2.3; Project # 3622-21000-029-00D)

<u>Transgenic manipulation of sorgoleone production in sorghum.</u> Sorghum plants produce a compound referred to as sorgoleone that could have beneficial properties for fighting weeds. ARS researchers in Oxford, Mississippi, have uncovered critical genes required for the synthesis of this compound, and are manipulating its levels in sorghum plants by modifying the expression of one of these genes. Such manipulation could be very useful in improving the weed-fighting capacity of sorghum by increasing sorgoleone production. Conversely, knocking out sorgoleone production could reduce problems encountered by farmers planting other sensitive crop species in the same field immediately following a sorghum crop, thus facilitating more efficient crop rotations. (NP302; C2, PS 2C; PM 2.2.3; Project # 6408-21410-006-00D)

Control network governing early crop seedling establishment and photosynthesis deciphered. Seedling establishment is an important trait that impacts crop production globally as well as adaptation to global change. However, the genetic and molecular determinants for improving seedling establishment are not well defined. At germination a seed must activate dormant pathways, and commence vigorous growth in darkness toward the soil surface, where upon exposure to light the photoreceptor phytochrome promotes a second transformation into the photosynthetically active state. ARS scientists in Albany, California, revealed that light activated phytochrome binds to a cohort of PIF (Phytochrome-Interacting Factors) proteins inducing their rapid degradation, thus releasing genes responsible for photosynthesis and green seedling growth. This research establishes a foundation of basic information for the genetic determinants for improving crop stand establishment in target crops. (NP302; C1, PS 1A; PM 2.2.3; Project # 5335-21000-027-00D)

Discovery of a master regulator gene involved in barley disease defense. Cell death is a key component of disease resistance. Hence, new information about the molecular mechanisms of cell death can provide insight into how to protect plants from disease. ARS researchers in Ames, Iowa, have used a combined strategy of high-throughput transcript profiling with classical genetic analysis of cell death mutants to identify a barley gene that regulates cell death. The gene encodes a key ribosomal RNA processing protein, so the results suggest a role of rRNA processing genes in mediating plant defense responses. The gene appears to be a master regulator that controls the expression of dozens of other genes, which are necessary for plant survival, stature, and yield. This discovery, also supported by the National Science Foundation-Plant Genome Research Program, provides new information that can be exploited by plant scientists in designing more effective mechanisms to protect plants from disease. (NP302; C1, PS 1B; PM 2.2.3; Project # 3625-21000-049-00D)

Development of a method for selecting soybeans with reduced levels of a major protein allergen. Allergenic proteins can reduce the nutritional qualities of soybeans used in food and feed applications. ARS scientists in Columbia, Missouri, discovered that some soybean lines lack a major allergen because of a specific mutation in the gene encoding the protein. A method was developed to directly select for the mutant gene. Adoption of this method will accelerate the rate at which new soybean varieties containing this trait can be evaluated for improved nutritional properties and human health. (NP302; C2, PS 2C; PM 2.2.3; Project # 3622-21000-029-00D)

A broccoli gene that impacts nutritional content in vegetable crops. Selenium is an essential trace mineral. Both biosynthesis and volatilization of selenium compounds affect the accumulation of the bioactive forms of selenium in crop plants. Broccoli accumulates high level of bioactive forms of selenium. To reduce selenium volatilization for producing healthy and more nutritious crops, ARS scientists in Ithaca, New York isolated a novel broccoli gene whose product mediates selenium volatilization, and utilized this knowledge to manipulate the nutritional value of crops via reduced nutrient loss due to natural volatilization. This discovery opens up new avenues toward increasing the accumulation of bioactive compounds in plants. (NP302; C1, PS 1B; PM 2.2.3; Project # 1907-21000-025-00D)

RNA controls seed production. Seed production and plant development are initiated by a double fertilization event during pollination that triggers both embryo and endosperm development. The coordination of these processes is highly susceptible to stresses associated with global change, including heat and water stress. ARS scientists in Albany, California, are genetically dissecting the cell and molecular processes responsible for the coordination of fertilization in plants. Genetic and molecular studies revealed that in male sperm cells, two genes work together to produce a natural double-stranded RNA molecule (nat-siRNA), that down-regulates the activity of a control gene that functions specifically in sperm. Plants whose sperm cells lack this important nat-siRNA rarely complete double fertilization and hence seed set is reduced. This discovery established a new paradigm for the role of nat-siRNAs in

coordinating plant development and seed production. (NP302; C1, PS 1A; PM 2.2.3; Project # 5335-21000-030-00D)

Tracking peanut gene expression patterns. To sequence the genome of the peanut, a comprehensive tool for dissecting its complex genetic mechanisms is needed. ARS scientists in Lubbock, Texas, developed a tool to analyze changes in the expression of 49,205 peanut genes and tested the utility of this tool on a variety of peanut tissues. This is the first large-scale, publicly available tool for determining which specific peanut genes are active out of all of the genes that exist within the peanut plant. The results generated by this tool will provide starting points for in-depth studies on finding candidate genes that can be utilized in reverse genetics to assign gene functions and identify specific molecular mechanisms of peanut response to environmental signals, developmental stages, and yield quality characteristics. (NP302; C2, PS 2A; PM 2.2.3; Project # 6208-21000-016-00D)

Overexpression of a blueberry gene protects against freezing. Losses in fruit quality and yield due to freezing can be devastating in blueberries. ARS scientists in Beltsville, Maryland, cloned and sequenced a blueberry gene called CBF that is important because it is responsible in many species for turning on genes that confer cold tolerance. CBF was introduced into blueberry and other species and was shown to increase freeze tolerance in non-acclimated plants. This research enables the development of new berry varieties with increased tolerance to cold through the use of molecular breeding approaches. (NP302; C1, PS 1A; PM 2.2.3; Project # 1275-21000-180-00D)

NP305

Commercialization of varroa-resistant honey bees selected for pollination performance. Varroa mites are an external parasite of honey bees and the major cause of colony losses throughout the United States. Bees with varroa sensitive hygiene (VSH), which have good resistance to varroa mites, were tested by ARS researchers in Baton Rouge, Louisiana for two seasons in a commercial migratory beekeeping operation focused on crop pollination. Bee colonies were created from VSH queens which were outcrossed, i.e., matings were not controlled, a method used by most large scale beekeepers. Bee colonies were shipped nationwide and used for spring pollination of almonds in California, apples in New York, low bush blueberries in Maine, and cranberries in Massachusetts, as well as late summer honey production in New York. VSH bee colonies performed well in terms of survival, populations, and resistance to varroa mites. The best surviving VSH bee colonies from each year were propagated to form a breeding population which had enhanced genetics for both mite resistance and behavior related to crop pollination. These bees are now being marketed by a CRADA partner (Glenn Apiaries). The use of their germplasm should improve adoption of mite-resistant bees by commercial beekeepers that pollinate crops. (NP305; C2, PS A2; PM 4.2.4; Project # 5428-21000-013-00D)

Brown marmorated stink bug can cause significant damage to apple and peach. The brown marmorated stink bug is a recently introduced invasive pest in eastern North America that has begun to cause economic damage and is a household nuisance. ARS scientists in Kearneysville, West Virginia, showed that early season damage is greater than 20 percent in apple and 50 percent in peach; in both unsprayed and sprayed orchards. The insect growth rate was also confirmed. Damage estimates and sampling will enable stink bug monitoring for determination of when damage levels to fruit are imminent. Application of these data to previously developed growth rate models will predict when the brown marmorated stink bug is at the proper growth stage to manage. (NP305; C1, PS 1B1; PM 2.2.3; Project # 1931-21000-019-00D)

<u>Leaf reflectance provides an estimate of sugarcane sucrose levels</u>. Current methods used to estimate sugarcane stalk sucrose levels prior to harvest are labor and time intensive. ARS scientists at Houma,

Louisiana; collected reflectance data from the leaves of predominant sugarcane varieties that were also sampled throughout two harvest seasons to determine sucrose accumulation (maturity). Leaf reflectance was effective at predicting sucrose in 36 to 79 percent of the cases, if varieties were combined; and in 65 to 100 percent of the cases if the varieties were considered separately. Several spectral regions were identified that appeared to be important in describing stalk sucrose levels, including: ultraviolet, blue, and green, and yellow, orange, and red, and the near-infrared wavelengths. These combined results indicate that it may be possible to utilize remote sensing techniques to estimate sugarcane maturity prior to harvest, which would allow growers and mills to more effectively manage field and varietal harvest schedules to insure maximum sucrose yields. (NP 305; C1, PS 1B2; PM 2.2.3; Project # 6410-12210-001-00D)

New plant acid-based varroa mite treatment developed. ARS scientists in Tucson, Arizona created a formulation using plant acids that is highly effective in reducing varroa mite populations in bee colonies. These plant acids are food grade compounds and are on the FDA's "generally recognized as safe" list. The product delivery system causes bees throughout the colony to have levels of the product that result in varroa mite mortality in less than 48 hours which does not cause mortality in either adults or immature life stages nor disrupt queen egg laying or colony growth. The product does not accumulate in the wax comb and, in most cases, was not found in honey samples; when it was found it was in very low amounts, less than 100 parts per billion. The product was developed under a CRADA and is in commercial production under the name "HopGuard." This product should significantly improve mite control in a manner that is non-toxic to the bees. (NP305; C2, PS A1; PM 4.2.4; Project # 5428-21000-013-00D)

Hand-held machine for assessing two-spotted spider mite damage in cotton. The two-spotted spider mite is an important pest of cotton and many other field crops. Early detection of plant damage caused by the mite is difficult because initial infestations tend to be scattered in small areas in the field. ARS researchers in College Station, Texas, adapted a hand-held light-reflecting (multispectral) instrument to detect the mite in growing cotton and beans. The instrument is capable of reliably distinguishing mite infested plants from non-infested plants, and is capable of differentiating between light, medium, and heavy spider mite infestations on cotton. This new technology will be very useful in detecting early spider mite infestations in cotton and other crops, and will guide rapid-response control procedures to assure effective protection of crops from mite damage; using the lowest amount of pesticide possible, with a minimum of adverse environmental impacts. (NP305; C1, PS 1A1; PM 2.2.3; Project # 6202-22000-028-00D)

Plants broadly attractive to bees needed in restoration of rangelands damaged by fires. Wild bee communities provide pollination services critical to large post-fire restoration projects in rangelands, but their fates following fires were largely unknown. ARS scientists in Logan, Utah, found that fires in rangeland plant communities that were in good shape before burning did not destroy native bees because the bees nest below ground; allowing them to rebound the next year. However, wildflowers which the bees depend on may be gone after a fire, and without them the bees cannot survive. This work demonstrates the importance of initial seeding in restoration projects. Initial seedings should include plants that are broadly attractive to local native bees in order to sustain these surviving bees until the native plant communities can re-establish, which can take several years. This work will be used by land manager in restoring habitat. (NP305; C2, PS 2B2; PM 2.2.3; Project # 5428-21000-013-00D)

Transition of alternative switchgrass substrate from replicated research to adoption by nursery producers. Pine bark is currently used as the primary potting substrate for the nursery industry, but its cost is increasing, its availability decreasing, and it must be transported from the southern United States over long distances. ARS scientists in Wooster, Ohio, have developed a new potting substrate comprised primarily of switchgrass, a biofuel crop that can be grown and harvested locally. The goal was to apply the research findings to a commercial scale operation. The researchers obtained a large quantity of

switchgrass, used a cooperating commercial nursery's equipment and traditional production procedures to pot plants with the new substrate, and compared the process and outcomes to that of traditional pine bark-based substrates. The switchgrass substrate performed as well as the traditional pine bark-based nursery substrate, demonstrating it as a viable alternative. Successful adoption of switchgrass substrate could decrease the industry's reliance on pine bark that must be transported from far distances, significantly lowering substrate cost. (NP305; C1, PS C4; PM 2.2.3; Project # 3607-21000-014-00D)

Nesting methods successfully established for four bumble bee species. Bumble bees are important pollinators of commercial greenhouse crops, but are difficult to rear in captivity. Because it is particularly difficult to get queens to establish new nests in captivity, ARS scientists in Logan, Utah, tested three nest establishment methods on four species of bumble bees. Results show that some techniques greatly increased the success rate (four-fold) in some species, in comparison to other techniques. By targeting proper rearing techniques to a given species, producers and researchers will be able to save time and resources when producing bumble bees in captivity. (NP305; C2, PS B2; PM 4.2.4; Project # 5428-21000-013-00D)

Grapevines are far less vulnerable to water-stress induced formation of vapor bubbles than previously reported. Vulnerability to drought-induced formation of vapor bubbles was evaluated in grapevines using Nuclear Magnetic Resonance in in-situ imaging of live grapevines to track movement of, and subsequent vessel blockage by, vapor bubbles. ARS scientists in Davis, California, revealed grapevines are not susceptible to significant drought induced vessel blockage within normal operating water potentials. The researchers also used High Resolution Computed Tomography, a type of CAT scan, to model the xylem network of grapevines. Using unprecedented resolution of this imaging technology, the researchers were able to describe the mechanism of air bubble blockage repair in grapevines. Researchers worldwide had suspected this mechanism for decades, but had been unable to visualize this process in living plants. (NP305; C1, PS 1B3; PM 2.2.3; Project # 5306-21220-004-00D)

New application procedures increase herbicide efficacy. The wide use of the herbicide glyphosate (Round-Up) on crops can create a situation where target weeds begin to develop a resistance, thus making the herbicide less effective. When this happens, it can require applicators to use more and more glyphosate in their treatments, which increases costs to both applicators and farmers and further exacerbates the weed resistance phenomenon. To address this growing problem, ARS researchers in College Station, Texas, studied how spray droplet size affects glyphosate effectiveness. The work showed that a higher spray droplet density (number of droplets per given area) results in better weed control. This knowledge allows applicators to adjust their application equipment in a manner to achieve good weed kill while using significantly less glyphosate and resulting in significant cost savings. An added benefit is that using less glyphosate to achieve the desired result also serves to slow down the process of weeds developing glyphosate resistance. Increasing the useful lifetime of an important and environmentally friendly herbicide like glyphosate also pushes into the future the need to shift to more expensive, environmentally harsh chemicals for weed control. (NP305; C1, PS 1A2 & 1B2; PM 2.2.3; Project # 6202-22000-028-00D)

Food Safety (ARS Goal 4) (108)

Select Examples of Recent Progress:

<u>Toxigenic E. coli in produce growing areas in California.</u> Many outbreaks of fresh produce associated foodborne illness have been linked to California's Salinas Valley, which has been called "the salad bowl of the world." The initial source of the contamination in outbreaks of E. coli O157 linked to leafy greens

produced in the Salinas Valley remains unknown. There has been a need to establish a baseline for the environmental prevalence of E. coli O157 and non-O157 E. coli in this important agricultural region. In collaboration with the University of California, Davis, and the California/USDA Animal and Plant Health Inspection Service Wildlife Services, ARS researchers at Albany, California isolated over 3,000 strains of E. coli O157 and non-O157:H7 Shiga-toxin-positive E. coli from 12,000 samples from water, animals and their feces, crops, and soil, and have determined the pathogens' genotypes and virulence gene profiles. ARS has submitted data to the FDA and to the CDC PulseNet. Data indicates that O157 and non-O157 E. coli prevalence varies considerably among sources, with the highest prevalence associated with cattle feces (7 percent and 33 percent, respectively), but that other animal species are also a significant source of the pathogens. This information, which provides the industry and public health agencies with the first epidemiological data for E. coli in the Salinas Valley, will be used to develop good agricultural practices for produce production.

(NP108, C 1.2, P.S. 1.2.3, P.M. 4.1.2, Project # 5325-42000-044-00D)

Assays for ricin and clostridium botulinum toxins. The potential use of ricin and botulinum neurotoxins as bioweapons in foods highlights the necessity for developing detection methods that work well for food samples. ARS scientists in Albany, California validated a new ARS method for the detection of ricin in economically important food matrices. The method exploits the specificity of antibodies with the enormous amplification provided by the polymerase chain reaction (PCR) technique, to enable measurement of about one billionth of a gram of this toxin in a golf ball sized portion of food. ARS scientists also developed a sensitive test for botulinum neurotoxin serotype B, the second most common form of this toxin. The test uses new monoclonal antibodies developed in the Albany laboratory and could detect less than one billionth of a gram of toxin in a teaspoonful of milk. This neurotoxin assay is 50 times more sensitive than the standard mouse bioassay. These two technologies could be used in regulatory laboratories by investigators seeking the source of foodborne contaminants and by the DHS and related agencies to assure the safety of the food supply. (NP108, C 1.2, P.S. 1.2.1, P.M. 4.1.2, Project # 5325-42000-043-00D)

Mycotoxin biological control agents native to Africa. Aflatoxins are potent fungal toxins that frequently contaminate foods. Improved methods to prevent aflatoxin contamination are needed particularly in the United States and Africa. To date, the most successful strategy for limiting aflatoxin contamination of crops is a biocontrol where atoxigenic strains of Aspergillus flavus (strains that lack the ability to produce aflatoxins) competitively exclude aflatoxin producers from environments where crops are grown. In the U.S., atoxigenic strains native to North America are commercially available for aflatoxin management. In several African nations the staples, corn and peanuts, frequently are contaminated with aflatoxins; humans there consume unsafe aflatoxin levels. ARS researchers in Tucson, Arizona, in collaboration with colleagues at the International Institute of Tropical Agriculture, Nigeria and the University of Arizona have selected atoxigenic strains of Aspergillus flavus useful in biological control. Tests of the Nigerian strains in farmers' fields in Nigeria demonstrated excellent efficacy at reducing contamination. The research demonstrates that effective atoxigenic strains can be isolated even from areas with severe contamination. The identified strains are a potential resource for reducing human exposure to aflatoxins in both East and West Africa. Advances made in biological control in Africa will help optimization of biocontrol in the U.S. and serve to improve food safety and security worldwide. These scientific endeavors are strongly supported by various agencies within the USDA, the Gates Foundation, the European Commission, and the World Health Organization (WHO). (NP108, C 2.1, P.S. 2.1.5, P.M. 4.1.1, Project # 5347-42000-019-00D)

<u>Soil solarization on cattle feedlot pen surfaces</u>. Soils at the feedlot pen surface are a source for transmission of Escherichia coli O157:H7 and a target for preharvest control measures to reduce this pathogen in cattle. Solarization is a pre-planting pathogen and pest control technique used in food and ornamental crop production that utilizes solar energy to heat the soil. ARS scientists at Clay Center,

Nebraska, determined that soil solarization is effective for reducing E. coli O157:H7 and generic E. coli from feedlot pen surfaces. Generic E. coli levels were reduced by 99.9% after 10 weeks of soil solarization but remained unchanged in unsolarized soils. E. coli O157:H7 was no longer detectable by 8 weeks of solarization, but could still be detected in unsolarized soils at 10 weeks. Use of soil solarization if implemented by industry will have an immediate impact through reducing the transmission and persistence of pathogenS among cattle and the production environment. (NP108, C 1.1, P.S. 1.1.4, P.M. 4.1.1, Project # 5438-32000-026-00D)

Too much disinfectant is a bad thing. Disinfectants are routinely used in the commercial rearing of livestock and poultry as a means of keeping facilities clean and healthy for both animals and the personnel working in the facilities. Very little is known about how pathogenic or food-poisoning microorganisms associated with animals and their surroundings can become resistant to disinfectants. ARS researchers at College Station, Texas, and Clay Center, Nebraska, showed that a significant percentage of the Escherichia coli O157:H7 bacteria isolated from living cattle, from cattle processing plants, and from ground beef were resistant to the effects of some common disinfectants used in cattle rearing facilities. The work also showed that if disinfectant used in the animal facilities was applied at higher than recommended levels, E. coli resistance was even more of a problem. The study's impact for the cattle industry was important because it showed that disinfectants must be used wisely in animal production facilities so as to assure that their positive effects are not offset by negative effects, which can include causing certain harmful bacteria to become even more problematic in animal production and human food safety. (NP108, C 1.1, P.S. 1.1.5, P.M. 4.1.1, Project # 6202-32000-020-00D)

Microarray detection of antibiotic resistance. Analysis of antimicrobial resistance genes in bacteria is a critical component in understanding the development and transmission of antibiotic resistance in humans, animals and the environment. ARS scientists at Athens, Georgia, developed microarrays for the detection of multi-drug resistant (MDR) plasmids in Salmonella and E. coli. The DNA microarrays were constructed containing probes for ~500 genes commonly found in different plasmid types. This technique should allow the rapid analysis of any genetic elements responsible for widespread MDR. The microarrays are being used to examine clinical isolates from animals collected by the National Veterinary Services Laboratory (NVSL), and through the National Antimicrobial Resistance Monitoring System (NARMS). These studies will allow monitoring by Federal agencies and researchers of the spread of MDR plasmids and determine their threat to human and animal health. (NP108, C 1.1, P.S. 1.1.5, P.M. 4.1.1, Project # 6612-32000-002-00D)

Chicken breeding for resistance against pathogens. Commercial poultry can be infected by bacteria and other microbes that cause disease in the birds, or that can cause food borne illness in humans who consume contaminated poultry products. The development of new strains of poultry that are inherently resistant to infection and colonization by Salmonella and other harmful bacteria would be a major accomplishment. ARS researchers at College Station, Texas, have for several years been working with an industry cooperator to develop such birds. The work has now progressed to the point that a separate, resistant population of breeder chickens has been developed, and these birds retain all desired productivity and quality traits (weight gain, feed conversion efficiency, meat quality, etc.). The resistant birds have much more efficient immune systems, key to fighting off infection by Salmonella and other pathogenic microorganisms, including the protozoan parasite (Eimeria) that causes the very destructive poultry disease coccidiosis. The accomplishment establishes, for the first time, that conventional breeding approaches can be used to develop birds that have a highly efficient natural immune system such that they will not become infected with harmful microorganisms; it is exceptionally important to efficient and profitable poultry production, to minimization of antibiotic usage, and to the provision of microbiologically safe food for the consumer. Expected commercialization of pathogen-resistant poultry lines, and their widespread utilization by producers, will in a very real sense revolutionize poultry production in the United States. (NP108, C 1.1, P.S. 1.1.3, P.M. 4.1.1, Project # 6202-32000-021-00D)

Antimicrobial packaging inactivates bacterial pathogens. Beverages can occasionally become contaminated by the bacteria Escherichia coli O157:H7 and Listeria monocytogenes, leading to foodborne illness outbreaks and product recall. To reduce the potential for growth of these pathogens in liquid products ARS researchers at Wyndmoor, Pennsylvania, developed antimicrobial packaging films and coatings for pathogen control in strawberry purees, milk and liquid egg products. Generally Recognized As Safe (GRAS) antimicrobial compounds, singly and in combinations, were incorporated into polylactic acid polymer films or coated on glass jars. The antimicrobial treatments inactivated more than 99.99 percent of E. coli O 157:H7 in strawberry purees and completely inactivated L. monocytogenes in milk and liquid egg white. The new technology if approved by the Food and Drug Administration and implemented by industry has the potential to significantly reduce foodborne illness outbreaks and product recalls. (NP108, C 1.2, P.S. 1.2.4, P.M. 4.1.2, Project # 1935-41420-013-00D)

Food processing costs. Novel non-thermal intervention technologies such as high pressure processing (HHP) and pulsed electric fields (PEF) processing are being developed to produce safer and more nutritious food. Research has shown the benefits of these technologies; however, there are no studies comparing the costs to that of traditional thermal (heat) processing. An analysis was performed, by ARS researchers in Wyndmoor, Pennsylvania, on the costs to process orange juice by these three different technologies. Results showed that thermal pasteurization was 4 cents/kg and 5 cents/kg cheaper than PEF and HPP, respectively, based on a 3,000 L/h production scale. This information is important to food manufacturers since, although non-thermal technologies offer benefits to the consumer in terms of improved quality, they are more expensive than thermal processing. Increased processing costs can place severe economic constraints on industry which are ultimately transferred to the consumer. (NP108, C 1.2, P.S. 1.2.4, P.M. 4.1.2, Project #1935-41420-013-00D)

Microwave with feedback technology. Basic microwave ovens heat food quickly and efficiently, but do not brown or bake food in the way conventional ovens do. This makes them unsuitable for cooking certain foods. Food safety regulatory agencies have expressed safety concerns relative to cooking/time temperature combinations for some poultry products, and cold spots in the products that could lead to insufficient cooking. A temperature-controlled microwave oven was used by ARS researchers at Wyndmoor, Pennsylvania, for in-package pasteurization of a simulated product containing raw chicken breast meat and gravy. This microwave heating system was modified from a commercial inverter-based microwave oven and equipped with an infrared sensor and a data acquisition system. The oven was used to cook chicken meat in gravy inoculated with strains of Salmonella. The study demonstrated that Salmonella inoculated onto raw chicken meat in the simulated product can be killed with a proper selection of heating strategies. This work will assist the USDA Food Safety and Inspection Service and the Food and Drug Administration to develop new time and temperature requirements for microwave cooking of poultry. (NP108, C 1.2, P.S. 1.2.4, P.M. 4.1.2, Project # 1935-42000-054-00D)

Not all Salmonella are created equal. Salmonella is a major cause of foodborne illness often caused by the consumption of contaminated poultry. Chickens from commercial processing plants are found most often to be contaminated with one of two types of Salmonella: S. Typhimurium or S. Kentucky. While both types may be found S. Kentucky is rarely found in human clinical cases of foodborne illness, whereas S. Typhimurium is often the cause of the illness. Studies by ARS scientists in Princess Anne, Maryland, found that S. Kentucky grows much slower on chicken than S. Typhimurium, which may explain why it rarely causes illness in humans. The researchers also developed a computer model that predicts the difference in risk to humans of chicken contaminated with these two types of Salmonella. The model was transferred to the USDA Food Safety and Inspection Service and industry through the ARS Pathogen Modeling Program and Combase. The impact of the work is projected to save the chicken industry and consumers millions of dollars per year by better predicting chicken safety. (NP108, C 1.2, P.S. 1.2.7, P.M. 4.1.2, Project # 1935-42000-057-00D)

Detecting and sorting mycotoxin contaminated grains. Processors and consumers in third world countries would benefit from being able to efficiently screen mycotoxin contaminated grains. Consumption of mycotoxin contaminated grain are serious food safety and food security concerns as contamination causes both acute (death) and long-term sequelae (cancer) in infected persons. ARS scientists in Peoria, Illinois, evaluated the performance of a rapid single kernel sorter that receives near-infrared reflectance spectra (NIRS) from kernels tumbling down a light tube and color image based sorting devices recently developed by ARS in Manhattan, Kansas, to discriminate corn kernels infected by eight fungus species at different levels of infection. Discrimination was done according to the level of infection and the mold species. In general, both instruments have good recognition of heavily infected and uninfected foods. Classification accuracy of the color imaging system was less, although the instrument has a lower cost and a higher throughput rate of approximately 75 kernels/s per channel or 40 Kg/hr. Systems using solar power or other green energy sources are being considered for development. The impact of developing a grain sorter that could be used in third world countries would be enormous, improving the health, safety and security of these populations. (NP108, C 2.1, P.S. 2.1.1, P.M. 4.1.2, Project # 3620-42000-033-00D)

Salmonella in dairy cattle. Salmonella contamination of milk is a critical issue for the Food and Drug Administration (FDA), as some States allow producers to sell unpasteurized milk and milk products. ARS scientists in Beltsville, Maryland, partnered with several universities to evaluate fluctuations in Salmonella serotypes on dairy farms in the Northeast U.S. Chronic shedding and population shifts are common in dairy cattle and longitudinal projects provide the opportunity to understand the dynamics of the interactions between serotypes, management strategies, and herd/cow prevalence, in order to identify and measure potential intervention or control measures. This research has identified significant shifts in serotypes and sources of the introduction of various serotypes. The studies have also provided an opportunity to evaluate a novel vaccine developed by ARS to control Salmonella Newport infections and to measure its effect on other serotypes. This collaborative study is providing the ability to identify potential management practices that are risk factors for foodborne outbreaks, and the ability to measure the impact of various pre-harvest interventions for foodborne pathogens in dairy cattle. This information is useful to both the regulatory agencies (FDA and USDA-FSIS), and to industry. (NP108, C 1.1, P.S. 1.1.2 and 1.1.4, P.M. 4.1.1, Project # 265-32000-078-00D)

Organic production system. Organic production systems are one of the fastest growing segments of U.S. agriculture for over a decade with an average increase of 15-20%. Yet, organic poultry production has unique challenges; the lack of safe, approved, and effective treatments for diseases that can adversely affect bird health and the wholesomeness of poultry products. ARS, in collaboration with the University of Arkansas, has developed a state-of-the-art organic/pasture poultry research facility. The facility meets the requirements of the USDA National Organic Program, the National Organic Standards Board and the Organic Poultry Guidance Document of the Accredited Certifiers Association. This facility is one of the very few organic certified poultry research facilities in the United States. It is the only facility, to our knowledge, in which both large-scale and small-scale production can be studied, and will provide the facilities to conduct important food safety research in organic production. (NP108, C 1.1, P.S. P.M. 4.1.1, Project # 6226-32000-009-00D.

Broiler house lighting affects Salmonella in poultry. The intensity of lighting, and also the light/dark cycle (total hours in light and dark during a 24-hour period) are factors used in commercial broiler growout to ensure bird health and improve production (growth rate, etc.). There is limited knowledge about the effect of lighting on general bird health and on the microbial ecology in the bird gut. There is limited information on how lighting might affect how Salmonella colonizes or otherwise affects the growing birds. ARS researchers at College Station, Texas, working with colleagues at Mississippi State University, showed that different lighting protocols do in fact affect Salmonella colonization of the birds. This finding is important because it indicates that appropriate lighting protocols can reduce the Salmonella problem. If such protocols are confirmed to be compatible with necessary production

parameters, they can be easily and cost effectively implemented by industry to produce microbiologically safer birds. (NP108, C 1.1, P.S. 1.1.4, P.M. 4.1.1, Project # 6202-32000-024-00D)

Livestock Protection (ARS Goal 4) (103, 104)

Select Examples of Recent Progress:

NP103

Babesiosis and horses. Equine piroplasmosis is a disease caused by blood parasites of the babesia family. It is considered a foreign animal disease in the United States and every effort has been made to prevent its entry into the U.S. horse population. The presence of this organism would prove very expensive to the equine industry due to blocking the export and importation of horses. During 2010 the U.S. encountered the reemergence of babesiosis, also known as piroplasmosis in the equine population. Babesiosis in horses is caused by two distinct parasites, *Babesia equi* and *Babesia caballi*. In order to begin developing strategies to control and re-eliminate the organism for the U.S. equine population and in response to the needs of the APHIS, scientists developed a method to eliminate persistent infection and transmission risk from horses infected with *Babesia caballi*. This has proven critical to the equine industry as it has resulted in owners of infected horses being able to treat their horses to enable them to resume their prior functions. In contrast the second parasite, *Babesia equi*, has proven more difficult to clear from infected horses. (N.P. 103, C 6, P.S.6B, P.M. 4.2.2, Project # 5348-32000-028-00D)

Foot-and-Mouth Disease Virus in cattle. It is well established that the respiratory tract is the most important route of infection of Foot-and-Mouth Disease Virus in cattle. However, conflicting data from different research groups have implicated regions of either upper respiratory tract (nasopharynx) or lower respiratory tract (lungs) as the primary sites of infection. Recent work by scientists at the Plum Island Animal Disease Center has demonstrated that after aerosol exposure to the virus the early pathogenesis events involve: (1) primary replication in epithelial cells of the pharyngeal mucosa-associated lymphoid tissue crypts and (2) subsequent widespread replication in pneumocytes in the lungs, which coincides with (3) the establishment of sustained viremia. This infection model demonstrated that massive viral amplification occurs in the lungs (with associated shedding to the environment) prior to appearance of the first vesicle. Viremia is established coincidently with further viral amplification in the lungs and at lesion (vesicle) predilection sites. This scientific information is critical to the development of Foot-and-Mouth Disease countermeasures as it is critical that prophylactic products target these previremic events in the pharynx and lungs. Based on this information, it is speculated that enhancement of mucosal immunity has a high probability of improving protection. Once viremia is established, on an individual animal basis, the battle has already been lost. Continued efforts to improve the understanding of virus host interactions during early phases of infection will greatly contribute to the development of effective tools to block viral infection. (N.P. 103, C 1, P.S. 1A, P.M. 4.2.1, Project # 1940-32000-052-00D)

Brucella suis infection in cattle. The elimination of Brucella abortus from cattle in the United States was the result of a national eradication program that began in 1934 with total costs exceeding 10 million dollars. Although several wildlife reservoirs remain a threat for reintroduction, ongoing monitoring activities are designed to detect any transmission events to domestic cattle. Although Brucella abortus has been largely eliminated from domestic cattle, the prevalence of Brucella suis in feral swine has emerged as a significant problem for domestic cattle. Brucella abortus infection of cattle is a regulatory issue and can lead to the loss of a State's brucellosis-free status and economic costs to producers and State regulatory agencies. Feral swine populations continue to increase in the U.S. and illegal transportation continues to expand their range into new States or regions. Contact with infected feral swine has led to

Brucella suis infections in a large number of cattle, particularly in the south and southeastern United States. Cattle infected with *Brucella suis* test seropositive (indicating previous exposure) on brucellosis surveillance tests and antibody responses cannot be readily differentiated from those due to infection with Brucella abortus. At this time, data is lacking on the time course for the antibodies to Brucella suis in the serologic responses of cattle after acute infection. In many States, vaccination for Brucella abortus using the vaccine RB51 is still utilized in domestic cattle. However, the efficacy of RB51 vaccination in protecting cattle against B. suis infection is unknown. In this study the serologic responses of cattle to Brucella suis infection and lesions and tissue localization of Brucella suis in pregnant RB51-vaccinated and control cattle after experimental challenge was assessed. The *Brucella* species remains an important disease in some areas of the United States. This study found that there was variation in the ability of various brucellosis screening or confirmatory tests in detecting infection of cattle with *Brucella suis*. Although experimental challenge with Brucella suis did not cause abortions in cattle, there was an increased incidence of retained placentas. In addition, cattle vaccinated with RB51 were not protected against Brucella suis infection. There was a high occurrence for Brucella suis to be found in the mammary gland with shedding in milk increasing the potential of transmission to humans. Research is ongoing to further develop vaccines strategies against *Brucella suis* in swine and cattle. (NP 103, C 3, P.S. 3A, P.M. 4.2.2, Project # 3625-32000-084-00D)

Arthropods and Rift Valley Fever virus. To determine which arthropods should be targeted for control should Rift Valley Fever virus be detected in North America, scientists in Manhattan, Kansas, in collaboration with the U.S. Army scientists at Fort Detrick, Maryland, evaluated mosquito species from the western, midwestern, and southern United States for their ability to transmit Rift Valley Fever virus. Culex tarsalis mosquitoes transmitted Rift Valley Fever virus efficiently. In contrast, when exposed to the same virus dose, none of the other Culex species tested transmitted Rift Valley Fever virus efficiently. Although Aedes vexans mosquitoes from Florida and Louisiana were relatively efficient vectors of Rift Valley Fever virus, specimens of this species captured in Colorado or California were virtually incompetent, illustrating the need to evaluate local population for their ability to transmit a pathogen. This is the first comprehensive vector competence study of U.S. mosquitoes for Rift Valley Fever virus and provides key targets for vector control should an outbreak of Rift Valley Fever virus ever occur in the United States. (N.P. 103, C 1, P.S. 1A, P.M. 4.2.1, Project # 5430-32000-001-00D)

Bovine tuberculosis eradication effort. Programs for eradication of bovine tuberculosis caused by Mycobacterium bovis from U.S. cattle were initiated in 1917, in large part due to public health concern of bovine tuberculosis as a zoonotic disease. Traditional test and slaughter policies have been effective in lowering the prevalence of disease, but the incidence of new cases has been increasing and efforts to eradicate bovine tuberculosis from the United States have been frustrating. A current problem impeding eradication of bovine tuberculosis from the United States is the persistence of Mycobacterium bovis infection in a reservoir of free-ranging white-tailed deer. This wildlife reservoir poses a serious threat to the bovine tuberculosis eradication effort. The Michigan Departments of Agriculture and Natural Resources and the Animal Plant Health Inspection Service, Veterinary Services have indicated that vaccines may be critical for the control of bovine Tuberculosis in Michigan. Scientists at Ames, Iowa, previously demonstrated that Mycobacterium bovis bacillus Calmette-Guerin used as a vaccine reduced disease severity in white-tailed deer upon experimental challenge with virulent Mycobacterium bovis. In an extension of these studies, scientists at Ames, Iowa, demonstrated that Mycobacterium bovis bacillus Calmette-Guerin persists in tissues of white-tailed deer for up to 9 months after vaccination. The attenuated live vaccine was primarily detected in lymphoid tissues without evidence of colonization of muscle (i.e., meat potentially consumed by humans); thus, the risk of transmission to humans is minimal. These findings underscore the necessity of continuing detailed safety studies for use of vaccines intended for wildlife that may be consumed by humans. (NP 103, C 3, P.S. 3C, P.M. 4.2.2, Project # 3625-32000-082-00D)

Monovalent vaccine for Swine influenza A virus. The gene constellation of the 2009 pandemic A/H1N1 virus is a unique combination from swine influenza A viruses of North American and Eurasian lineages, but prior to April 2009 this new and emerging virus had never before been identified in swine or other species. Although its hemagglutinin gene is related to North American H1 swine influenza A virus, it is unknown if vaccines currently used in U.S. swine would cross protect against infection with the pandemic A/H1N1. Scientists in Ames, Iowa evaluated the efficacy of inactivated vaccines prepared with North American swine influenza viruses as well as an experimental homologous A/H1N1 vaccine to prevent infection and disease from 2009 pandemic A/H1N1. All vaccines tested provided partial protection ranging from reduction of pneumonia lesions to significant reduction in virus replication in the lung and nose. The multivalent vaccines demonstrated partial protection, however, none were able to prevent all nasal shedding or clinical disease. An experimental homologous 2009 A/H1N1 monovalent vaccine provided optimal protection with no virus detected from nose or lung at any time point. Based on cross protection demonstrated with the vaccines evaluated in this study, the U.S. swine herd likely has significant immunity to the 2009 A/H1N1 from prior vaccination or natural exposure. However, consideration should be given for development of monovalent homologous vaccines to best protect the swine population, thus limiting shedding and the potential transmission of 2009 A/H1N1 from pigs to people. (NP 103, C 4, P.S. 4B, P.M. 4.2.2, Project # 3625-32000-088-00D)

Survey of cattle parasites and resistance to drug therapy. Intestinal parasites have been a problem for livestock for many years. Conservative estimates are that gastrointestinal parasites cost the American cattle industry over \$2 billion dollars per year. This cost is based on the treatment with chemicals (anthelmintics) to kill intestinal worms and the decreased productivity and growth of livestock infected with parasites. This easily makes gastrointestinal worms or nematodes the most costly parasitic infection of American cattle. Although the drugs currently used to control cattle intestinal worms worldwide are generally effective and safe, global resistance by parasites to drugs is rapidly on the rise. A national survey of cattle intestinal parasites and their response to anti-parasiticide treatment was conducted in collaboration with the APHIS and two university collaborators. The results of a study of randomly selected cattle operations demonstrated a wide distribution of resistance to anthelmintic treatment. In nearly all cases, the species of parasite was Cooperia species, in particular Cooperia punctata. While historically this parasite was a minor species infecting cattle, with the emergence of resistance to antiparasiticide treatment, it has become a predominant pathogen. These results demonstrate that overuse of anthelmintics has not only selected for drug resistant intestinal parasites, but has also changed the population dynamics of parasites on pasture and has resulted in a species with increased ability to cause damage to livestock. These results clearly demonstrate that there has been a rapid rise in the prevalence of cattle gastrointestinal parasites that are resistant to some of the most commonly used anthelmintics. Additional research is being conducted to assess potential genetic markers that can be used to identify anthelmintic resistant parasites which will aid in developing new intervention and control strategies to control important species of intestinal parasites in cattle. (N.P. 103, C 6, P.S. 6A, P.M. 4.2.1, Project # 1265-32000-082-00D)

<u>Direct-fed microbials improve the gut health in broiler chickens.</u> Probiotics modulate gut immunity and enhance natural resistance against avian coccidiosis. Direct-fed microbials are live microorganisms which confers a health benefit on the host by balancing its intestinal microbes. Recently, much attention has been paid to the role of direct-fed microbials on the immune system (i.e., immunomodulation) and their effects on the interaction between gut microflora and host immune system development. In order to develop a novel control strategy for poultry diseases and to reduce antibiotics uses, scientists in Beltsville, Maryland, investigated the immune mechanisms and possibility of immune enhancement using various direct-fed microbials products. Feeding dietary direct-fed microbials significantly improved intestinal structure and enhanced gut health as revealed by increased villus height and crypt depth compared with normal controls. These studies provided a rational scientific basis for future studies to investigate direct-

fed microbials as immunopotentiating agents to enhance host protective immunity against enteric pathogens in broiler chickens. (N.P. 103, C 2, P.S. 2C, P.M. 4.2.2, Project # 1265-32000-075-00D)

Glässer's disease in swine. Respiratory disease remains one of the most important causes of disease to the swine industry. Haemophilus parasuis is a bacterium that causes Glässer's disease in swine, a disease characterized by chronic debilitation and often death that costs the swine industry millions in losses annually. However, not all strains of the bacterium cause disease. To date, little is known about genetic differences among Haemophilus parasuis strains and genetic factors that contribute to its ability to cause disease. Respiratory and systemic diseases caused by Haemophilus parasuis remain problematic to the swine industry, especially high health status herds. Currently, there are no effective vaccines for Haemophilus parasuis and most control strategies have not been successful in preventing disease and the resulting loss of pigs. Scientists in Ames, Iowa, compared four isolates of the bacterium for their ability to cause disease in pigs. Three of the isolates caused disease in the pigs, while pigs given the fourth isolate remained healthy. Pigs that were given the non-disease causing isolate were subsequently protected from disease when exposed to one of the disease causing isolates. Thus, strains of *Haemophilus* parasuis that don't cause disease may be useful as vaccines to protect pigs against disease causing strains. In order to better understand the differences between *Haemophilus parasuis* isolates and potentially identify the mechanism by which some isolates fail to cause disease, DNA sequencing is being used to compare the genomes of the different isolates. These results will provide information so scientists can develop control strategies and potentially identify vaccine candidates that can be used by swine producers to control losses from this disease and identify whether strains of *Haemophilus parasuis* circulating on the farm will cause disease. (N.P. 103, C 4, P.S. 4B, P.M. 4.2.1, Project # 3625-32000-089-00D)

NP104

A potential savings by strategic releases of screwworm flies. The screwworm fly lays eggs in wounds and then burrows into the flesh of humans, cattle, and other mammals, often with fatal results. This horrific pest was eradicated from the entire continent of North America by systematic release of sterile males. A barrier of sterile males continuously released in Panama prevents reintroduction of the screwworm fly from South America. Scientists in Kerrville, Texas and Panama reviewed and analyzed release technologies to improve screwworm fly dispersal relative to where, when, and how many sterile flies are released in the barrier zone. Quantitative calculations were based on screwworm biology and modeling of results as well as application of global information systems. Recommendations with supporting data presented to the Panama – U.S. Commission for Eradication of Screwworm would: (1) result in updated equipment on-board dispersal aircraft, (2) strategically reduce the number of flies dispersed, and (3) save up to \$1 million in Federal funds annually. Recommended improvements to the navigational software and equipment are currently being implemented by APHIS' International Services and the Panama – U.S. Commission for Eradication of Screwworm. (NP 104, C 3, P.S. 3E, P.M. 4.2.2, Project # 6205-32000-035-00D)

<u>Vaccines to stop the cattle fever tick</u>. Cattle fever ticks are a worldwide pest of cattle that decimate the industry by the bleeding to death of animals, and by transmission of key diseases like babesiosis and anaplasmosis. These diseases not only reduce the efficiency of production, they also prevent trade in live animals. The cattle fever tick is a current threat to international food security and an imminent threat to the cattle industry in the southern United States where the tick was eradicated between 1907 and 1943. Scientists in Kerrville, Texas, collaborating with scientists from EMBRAPA (the Brazilian Agricultural Research Corporation) identified two anti-cattle tick vaccine candidates in cattle trials. These candidates had been prioritized in a prior ARS project through bioinformatic and molecular biological approaches. In the cattle trials conducted in Brazil, the candidates outperformed the recombinant Bm86 Campo Grande antigen which is an antigen similar to that used in the only current commercially available anti-

tick vaccine. An invention disclosure was filed; cattle trials are scheduled to evaluate various parameters in the vaccination protocol to optimize efficacy. The worldwide use of a consistently effective anti-tick vaccine in cattle would reduce production costs associated with tick treatment, increase the ability of under-developed countries to feed the hungry, and contribute to the maintenance of the eradication of cattle fever tick in the United States. (N.P. 104, C 3, P.S. 3A, P.M. 4.2.2, Project # 6205-32000-031-00D)

New methods for protecting US troops deployed overseas. Protection of deployed military from mosquitoes and flies that transmit disease is a critical component of successful military operations because U.S. military personnel are generally susceptible to the pathogens and they are more exposed to bites in the field. Scientists from Gainesville, Florida, carried out field trials of two insecticides (bifenthrin and lambda-cyhalothrin) as barrier treatments (applied to vegetation or to military materials between the source of biting arthropods and the location to be protected) in desert environments (Coachella Valley, California; western Kenya, Iraq, and Afghanistan) and in a humid sub-tropical environment in Florida. Results from vegetation treatments indicated significant reduction in mosquitoes in field counts and lab assays for up to a month; results from material treatments indicated significant reduction of mosquitoes or sand flies in field counts and lab assays for up to 18 months. Another way to reduce the number of vectors is application of aerosols of insecticide outdoors. Trials in a desert environment showed that thermal fog (machines that burn a mixture of insecticide and oil to produce a dense insecticide-laden smoke) was superior to ULV (ultra-low volume, which produces very small droplets in a mist that is made mechanically) and that malathion, an organophosphate, was better than sumithrin, a pyrethroid. The results suggest that, as an enhancement to the current Department of Defense pest management system (such as use of DEET and permethrin treatment of uniforms) barrier treatments may be successful in providing protection from vector-borne diseases for deployed troops in desert habitats by significantly reducing densities of mosquitoes or sand flies reaching individual personnel in protected areas. Thermal fogs of malathion were shown to be better than the usual treatment involving ULV pyrethroids. (NP 104, C 2, P.S. 2C, P.M. 4.2.2, Project # 6615-32000-045-00D)

Natural biological control of the imported fire ant. Fire ants, accidentally imported from Argentina in the early 20th century, are unusually abundant in the United States, because they have escaped their natural enemies left behind in South America. Scientists in Gainesville, Florida, obtained approval from the North American Plant Protection Organization and USDA-APHIS regulators to release a new species of phorid decapitating fly (Pseudacteon cultellatus) as a fire ant biocontrol agent. This new species of fly specializes on attacking the smallest sizes of fire ant workers, which are most abundant in multiple-queen fire ant colonies. This preference is especially important because multiple-queen fire ant populations average two to three times the densities of regular single-queen fire ant populations and are therefore a substantially greater pest of homes, agriculture, and the environment. Another example of significant progress in development of biological control was the comparison of a new fire-ant virus, SINV-3, from American and Argentinean populations. Working with the USDA ARS South American Biological Control Laboratory, scientists sequenced the SINV-3 genome in entirety from an Argentinean source and compared it with the genome sequence found in U.S. populations. The Argentinean variant had a different genomic architecture and may exhibit different virulence levels compared with the U.S. variant. ARS scientists also showed that more recent infestations of fire ants in Australia, China, and Taiwan probably originated from the United States rather than from South America. These subsequent invasions of fire ants may have been facilitated by genetic pre-selection in the United States. This knowledge is valuable because the same biological control agents that are effective in the United States are likely to be helpful in Asia and Australia. Establishment of a complex of biological control agents in the United States and other countries where the fire ant has invaded will likely establish the system of natural controls necessary to reduce the impact of this pest. (NP104, C 5, P.S. 5A, P.M. 4.2.2, Project # 6615-32000-044-00D)

Gene silencing, a novel method for mosquito control. Toxicants with new modes of action and high specificity are being investigated for mosquito control. Using gene silencing technology or RNA interference (RNAi), scientists in Gainesville, Florida have designed molecules that inhibit expression of critical proteins in mosquitoes that result in mortality. It was shown that these molecules can be delivered to adult mosquitoes through the cuticle, with other possible delivery methods under investigation. A new CRADA with industry will enable large scale production of RNAi molecules to investigate new carriers and delivery methods to mosquitoes. This technology will provide completely safe insecticides based on natural chemicals and will be easily modified for different purposes. (N.P. 104, C 1, 2, and 3, P.S. 1A, 2A, and 3F, P.M. 4.2.2, Project # 6615-32000-045-00D)

New ways to stop mosquito bites. Insect repellents applied to the skin are widely used to prevent mosquitoes from biting, but they are usually used as a last resort when other forms of control have failed. The promise of highly effective, easy-to-use repellents depends on a precise understanding of how these useful chemicals work. Scientists at Beltsville, Maryland, showed that insect repellents confuse mosquitoes in the manner that they stimulate the organs mosquitoes use to find humans. In the absence of other odors, the repellents activate the organs in a way that does not occur in nature, probably interfering with the ability of the mosquito to find a host. If odors are present that would usually attract the mosquitoes to a host, the repellents prevent detection organs from sensing the chemicals that would normally direct the mosquito to the host. The research demonstrates that repellents can have positive and negative effects on mosquito reception. After we understand how repellents work, we will be able to develop chemicals that work more precisely with the hope that we can manage mosquito behavior more effectively. Products could include more powerful insect repellents, spatial repellents that keep mosquitoes out of a home or backyard, and "herding" chemicals that put mosquito populations into areas where they either cause no harm or where they can be killed. (N.P. 104, C 1 and 2, P.S. 1a and 2b, P.M. 4.2.1, Project # 1275-32000-007-00D)

Mosquito immune response to a pathogenic virus. CuniNPV is a mosquito specific baculovirus that infects mosquito larvae within the genus *Culex*, which are important vectors of encephalitis viruses worldwide. Mosquito viruses have to overcome insect resistance mechanisms, just as human viruses have to overcome human immune systems. This work demonstrated the presence of a very unusual insect resistance mechanism and the virus' ability to overcome it. Basically, the virus appears to shut down the ability of the mosquito to modify its cellular architecture and the mosquito responds by overproduction of a chemical that regulates the process. Scientists in Gainesville, Florida, together with researchers at the University of Florida have investigated mosquito genes that are involved in the pro-apoptotic (programmed cell death) response to viral infection. Culex larvae were challenged with CuniNPV and the expression profile of the pro-apoptotic gene, mx, was measured. There was not a significant increase of mx expression before 8 hr post infection but the level of mx expression continued to increase throughout the infection period and at 48 hr post infection was about 10 times higher than the uninfected controls. The induction of mx did not result in apoptosis but rather necrosis (degradation of cells) indicating that CuniNPV prevents apoptosis despite the very high level of mx expression. It is possible that CuniNPV utilizes an as yet unknown, but powerful, mechanism to block the apoptotic pathway downstream of mx activation. Identification of this mechanism could have important implications on how other viruses in mosquitoes evade the host immune response, providing innovative new mechanisms for mosquito control. (N.P. 104, C 1 and 3, P.S. 1A and 3F, P.M. 4.2.1, Project # 6615-32000-045-00D)

Realistic assessment of mosquito behavior following exposure to insecticides and repellents. Control efforts against adult mosquitoes primarily consist of aerial spraying and residual treatments, usually using pyrethroids that affect the insects' nervous system. Insect repellents, often containing the active ingredient DEET, are used to reduce the number of bites from populations of mosquitoes that have otherwise escaped other control efforts. Not all mosquitoes contact a lethal dose of insecticide and the effect of sublethal exposure on sensory organs is poorly characterized. Scientists in Gainesville, Florida,

exposed female Aedes aegypti, Anopheles albimanus and Culex quinquefasciatus to LD25 levels of pyrethroid treatments (the dose necessary to kill only 25% of the mosquitoes). Mosquitoes treated with insecticides tended to fly a less direct route to an attractant and fewer of them responded to the attractant after 24 hours. This suggested that pyrethroids may have a greater impact on disease transmission than their immediate killing impact. Filming the flight behavior of host-seeking female Aedes aegypti and Anopheles in the presence of pyrethroid and DEET-treated uniforms, it was found that mosquitoes always detected the presence of the blood meals as indicated by their flight towards the blood meal, therefore, DEET did not mask the odors. However, both the DEET and pyrethroid treatment reduced the number of mosquitoes taking blood meals. Mosquitoes attempting to feed through the DEET were repelled and after 10 minutes the majority had stopped trying to feed. In the pyrethroid treatment, mosquitoes tried to feed throughout the entire 20 minute trial and this continued contact with the treated substrates often resulted in 10% mortality of the mosquitoes. Neither repellent nor insecticide created a spatial barrier or prevented mosquitoes from landing and attempting to feed through treated material. This work shows that permethrin and DEET have different effects, though both are used as repellents. Developing new active ingredients that affect mosquitoes in precise ways will lead to new ways to protect people from bites. (N.P. 104, C 1, 2, and 3, P.S. 1A, 2B, and 3F, P.M. 4.2.2, Project # 6615-32000-046-00D)

Cutting edge genomics applied to stable fly control. Scientists in Kerrville, Texas have used more streamlined methods of gene identification to find 21 genes associated with host seeking and egg laying in stable flies, including the first olfactory and taste receptors to be reported for this significant livestock pest. They were also able to find the specific change in a gene that results in resistance to the commonly used veterinary pesticide, permethrin. Through laboratory selection, University of Florida researchers previously obtained a 15-fold resistance to permethrin in a strain of stable flies. In collaboration with researchers in Gainesville, Florida an ARS scientist at Kerrville, Texas identified a mutation in the stable fly sodium channel gene that associates with the observed resistant phenotype. Among the many potential products from these discoveries are highly specific molecular pesticides, design of chemicals that alter stable fly behavior, and methods for field detection of insecticide resistance. (N.P. 104, C 3, P.S. 3B, P.M. 4.2.1, Project # 6205-32000-033-00D)

<u>Discovery of termite enzyme systems for pest control and bioenergy.</u> There are few molecular target sites for the development of new termite control chemicals. Scientists in New Orleans, Louisiana, have cloned two enzymes from the Formosan subterranean termite and expressed them in a bacterial expression system. Incubation of the two enzymes together with cellulose results in digestion of the cellulose to glucose. This will allow us to test a variety of possible inhibitory compounds against the digestive enzymes necessary for the termites as a possible wood preservative or termite control agent. An additional outcome for this research is the possible use of these enzymes for the conversion of biomass into biofuel. (N.P. 104, C 4, P.S. 4A, P.M. 4.2.2, Project # 6435-32000-012-00D)

Crop Protection (ARS Goal 4) (303, 304)

Select Examples of Recent Progress:

NP303

Mystery of the life cycle of wheat and barley stripe rust solved and protected wheat varieties released. Stripe rust (also called yellow rust) of wheat and barley causes significant wheat and barley crop losses worldwide but the life cycle of the rust fungus has long baffled scientists. ARS scientists in St. Paul, Minnesota have made the first identification of an alternate host for any type of stripe rust pathogen by demonstrating that several species of barberry serve as alternate hosts for the cereal rust. Stripe rust is

known to be one of the most variable cereal rust pathogens. This discovery suggests that recombination on susceptible barberry species is playing a key role in contributing to pathogen variability. This information can assist crop breeders in developing entirely new strategies to protect cereals from stripe rust losses. Another advance in cereal stripe rust protection was made by ARS researchers at Pullman, Washington who exploited stripe rust resistance assays and molecular markers to develop more than 10 new stripe rust-resistant wheat varieties. (NP303; C1&3, PS 1B, 3A & 3B; PM 4.2.4; Project # 3640-21220-020-00D and Project # 5348-22000-014-00D)

New varieties of table grapes and raisins with high fruit quality and Pierce's disease resistance. Introduction of Pierce's disease resistance from wild grape species into table grapes and raisins results in small berries and poor fruit quality. ARS scientists in Parlier, California have used traditional breeding techniques to generate desired *Vitis vinifera* table grape and raisin varieties that retain Pierce's disease resistance from the less desired grape, *Vitis arizonica*, but that have fruit of high vinifera quality. The current advanced selections will be developed into new table grape and raisin cultivars with Pierce's disease resistance. (NP303; C3, PS 3B; PM 4.2.4; Project # 5302-22000-008-00D)

Multiple approaches are being used to combat the citrus greening disease. It is difficult to cultivate the bacteria (*Candidatus Liberibacter asiaticus*) presumed to cause Huanglungbing (HLB) or citrus greening disease and in propagating and maintaining a large number of HLB-infected plants. Recent advances by ARS scientists in Fort Pierce, Florida include: identifying a combination of two chemicals that eliminates the pathogen associated with citrus HLB in the plant; developing a method for rapid detection of the presumed pathogen; and fully sequencing the genome of the presumed pathogen. These new developments will greatly facilitate HLB research and the development of new strategies for control of this devastating disease. (NP303; C1&4, PS 1B & 4A; PM 4.2.4; Project # 6618-22000-034-00D)

Identification of the causal agent associated with the almond brown line disease. Almond brown line disease was discovered in California in the 1990s as a graft union disorder in almonds grown on plum rootstock in orchards on marginal land. When the trees are infected by Peach yellow leafroll phytoplasma, a bacterium without cell walls, they develop a brown necrotic line at the graft union resulting in tree death. It has been difficult to prove the association of phytoplasma in infected almond trees because of the absence of a suitable detection assay. ARS scientists in Davis, California developed a molecular assay and successfully detected this phytoplasma in almond extracts. This assay will be used to monitor the trees in a commercial orchard impacted by almond brown line disease. (NP303; C4, PS 4B; PM 4.2.4; Project # 5306-22000-014-00D)

Mobile pathogenicity chromosomes identified that explains how harmless fungi can change into plant killers in wheat and barley scab. Keeping plants healthy is all about keeping one step ahead of the microbes that can harm them. Fungi that cause plant disease have an uncanny knack for changing and overcoming disease resistance genes that scientists work so hard to breed into plants. Fusarium species are among the most important pathogenic fungi and include *Fusarium graminearum* that causes wheat Fungal Head Blight or scab. Using comparative genomics, ARS researchers in St. Paul, Minnesota have determined that Fusarium fungal pathogens have the unusual ability to pass particular chromosomes between strains and convert harmless fungi into pathogens. This process explains why new strains of fungi can arise quickly and threaten crops. The discovery means scientists can now start working on a means to thwart the trading of chromosomes and the disease causing ability that comes with them. (NP303; C2 & 3, PS 2A & 3B; PM 4.2.4; Project # 3640-22000-023-00D)

A grower-friendly method for detection of Citrus tristeza virus. Citrus tristeza virus (CTV) continues to be a major limiting disease of citrus, and in California, is managed by state and federal regulatory agencies. The mandatory state eradication program has been replaced by a program of selective removal of trees that are only infected with the most severe strains, with the mild strain being far less destructive.

However, the state requires all citrus nurseries to propagate virus-free trees, regardless of the strain. ARS scientists in Parlier, California, in partnership with a private diagnostics company, developed a field-deployable, direct tissue blot assay to detect CTV. This is simple, sensitive, and cost-effective detection tool which nurseries can use to test their own trees, and has been used to successfully monitor CTV infection in thousands of budwood trees and nursery increase blocks. Infected trees were immediately removed by the nursery, thus, meeting the rigorous standard of maintaining and selling virus-free stock and propagations. Since CTV is readily aphid-transmitted, all citrus nurseries in California are vulnerable and can now use the testing kit for this same purpose. Before nurseries had to request regulatory agencies or the University of California to test their trees, which was expensive and time consuming. (NP303; C1, PS 1B; PM 4.2.4; Project # 5302-22000-009-00D)

Biocontrol agent for fire blight improved. Biological control of fire blight of apple and pear with beneficial microorganisms is a viable alternative to the use of antibiotics, which have become less effective due to resistance in the causal organism. One effective biocontrol agent, the bacterium *Pantoea agglomerans* strain E325, was originally discovered by ARS scientists in Wenatchee, Washington, and is now commercially available. In efforts to improve the production and tolerance of this organism to dehydration and related stress factors, salt levels were increased in the growth medium. As a result, the organism survived at significantly higher levels during freeze drying and subsequent storage prior to application, and in some cases, established better on floral (petals and sepals) surfaces under dry conditions. The research led to commercial modifications and an improved biological product for fire blight management. (NP303; C4, PS 4A; PM 4.2.3; Project # 5350-22000-015-00D)

Advancing bacterial pathogen recognition through genomics. Because of their small size and inability to grow outside living tissue, phytoplasmas (specialized plant pathogenic bacteria) are difficult to identify. ARS scientists in Beltsville, Maryland, have harnessed the power of genomics for plant bacterial pathogen detection. The USDA's Phytoplasma Classification Database, used worldwide by quarantine agencies, diagnostic companies, scientists, students, and faculty, was expanded significantly and made easier to use to aid in the rapid identification and classification of bacterial pathogens, including new species. A new Web-based tool, iPhyClassifier, for the online gene-based identification and classification of bacterial pathogens incorporates carefully curated databases of pathogen gene sequences for up-to-date classification and comparative studies. As a result, regulatory and quarantine agencies can detect and identify phytoplasmas more readily. This information is also being used to reduce crop losses through production of certified disease-free germplasm. (NP303, C1, PS 1B; PM 4.2.4; Project # 1275-22000-246-00D)

Root to root spread of the sudden oak death pathogen *Phytophthora ramorum* was documented under flooded conditions. When sprouted acorns were exposed to different concentrations of spores of *Phytophthora ramorum*, as little as one spore per milliliter caused infection within 24 hours in all four Eastern U.S. oak species tested. Forty-six plant species of economic or ecological importance to the Eastern U.S. were evaluated for their ability to support root infection and production of *P. ramorum* spores. Many of these species supported higher populations of the pathogen than a highly susceptible control species, *Viburnum tinus*. This information will be valuable for risk evaluation and regulatory decision-making. (NP303; C2, PS 2C; PM 4.2.4; Project # 1920-22000-036-00D)

Genetic Diversity in Reniform Nematode Populations Described Using Microsatellites. Understanding variability in the pathogen population is important not only to improve our understanding of the basic biology of this organism, but also to assist in tracking how the pathogen responds to various management practices and determine the origin and spread of the nematode in the United States. Using six populations of reniform nematode from four states, ARS researchers in Stoneville, Mississippi, developed 156 molecular markers that will enable detection of the genetic variability within this species. Twenty-two of these markers were sensitive enough to detect differences among three populations collected from the

same location in Mississippi. This information is being used by plant breeders to develop nematoderesistant plants. (NP303; C3&4, PS 3B & 4A; PM 4.2.4; Project # 6402-22000-005-00D)

NP304

Insect and mite systematics help safeguard the Nation's agriculture. Invasive species cause hundreds of billions of dollars in losses in the United States each year. Systematics collections are essential for addressing these threats. During the past year, ARS scientists in Beltsville, Maryland used these insect and mite collections to conduct 46,000 identifications, including over 6,000 considered urgent by APHIS, from specimens collected at U.S. ports. The researchers produced electronic identification tools for invasive fruit flies; descriptions of new parasitic wasps that attack leaf-mining flies and other wasps used for biocontrol of the invasive weed, Old World climbing fern; and identification of flea beetles used for biocontrol of other invasive weeds. In addition, this research produced additional knowledge on moths that is assisting in the "Discover Life in America" effort documenting life in the Great Smoky Mountains National Park. The scientists also are discovering clues to host-parasite evolution through leaf-mining fly systematics, and conducting extensive biological and ecological studies of a new parasitic wasp found on the important invasive emerald ash borer. These applications are being used to prevent the introduction of new invasive species and manage established ones. (NP304, C1, PS 1A; PM 4.2.4; Project # 1275-22000-230 [and 231, 232, 233, 236, 237, 238, 239, 255, 257, and 258]-00D)

New control solutions for aphids developed based on neuropeptide hormone technology. Pest aphids cause hundreds of millions of dollars of crop damage every year; many populations have already acquired resistance against insecticides used for control. ARS researchers at College Station, Texas, in cooperation with British colleagues, developed an entirely new approach for the control of pest aphids. The technology is based on developing versions of natural aphid hormones (known as neuropeptides) that resist metabolism (inactivation) by natural aphid body enzymes. Natural neuropeptides in aphids and other insects regulate critical life processes, such as water balance and digestion. Some of the neuropeptide "mimics" developed by this work match or even exceed the potency of current insecticides used in aphid control. While the development of commercially viable neuropeptide technology for aphid control has not yet been realized, this accomplishment is moving the research forward and is catalyzing related work by other scientists in industry, academia, and government. (NP304; C2, PS 2A; PM 4.2.3; Project # 6202-22320-002-00D)

Newly designed cover crop roller-crimpers results in minimal weed management inputs for soybean production. The use of zero-till systems for weed management continues to be a major goal in achieving sustainable organic crop production systems. ARS scientists in Urbana, Illinois have demonstrated that the use of newly designed cover crop roller-crimpers results in minimal weed management inputs for soybean production. This newly designed cover crop produces soybean yields similar to that of chemically terminated cover crops followed by post-emergent applications of the herbicide glyphosate. Other accomplishments by ARS scientists in Beltsville, Maryland demonstrated that the combination of early weed suppression by rye mulch, with the late weed suppression of densely sown soybeans, can provide high levels of soybean production. These results are leading to more effective and sustainable zero-till systems for organic cropping systems. (NP 304; C2, PS 2B; PM 4.2.4; Projects # 3611-22000-018-00D & 1265-22000-164-00D)

<u>Discovery and release of biological control agents of invasive species in Florida</u>. Florida has been hit hard by invasive species because of its subtropical climate. State and individual water management districts actively attempt to manage select invasive species of plants that threaten natural habitats using a combination of mechanical, chemical, and biological control, and through the development of natural enemies of invasive weeds and insect pests. In many cases, that involves finding biological control agents

in the native range of the invasive species. When biological control is successful, it solves the weed or pest problem without the addition of chemicals to the environment and sustains itself through the creation of a natural balance between species. ARS scientists at the Australian Biological Control Laboratory in Brisbane, Australia, the South American Biological Control Laboratory in Buenos Aires; Argentina, and the Invasive Plant Research Laboratory in Fort Lauderdale, Florida developed natural enemies against key invasive weed species in Florida. This included the establishment of a gall fly population, *Lophodiplosis trifida*, as a new biological control agent of the melaleuca paperbark tree; the discovery of a rove beetle that attacks skunkvine, an invasive weed that displaces native vegetation; and the completion of years of research to successfully release a new leafhopper biological control agent of waterhyacinth in Florida. These achievements will help preserve the native vegetation and wildlife in Florida at low cost and with minimal management in the future. (NP304; C2&3, PS 2A & 3B; PM 4.2.3; Projects # 0210-22000-004-00D & 0211-22000-006-00D)

Development of data to support the registration of pesticides for specialty crop uses. Growers of specialty crops such as fruits, vegetables, mint, hops, herbs, spices, and other minor acreage crops generally lack the pesticides that are available for major acreage crops such as corn, wheat, and other small grains, soybeans, and cotton. Pesticide manufacturers do not have the economic incentive to develop the data for labeling pesticides for their uses on these minor acreage crops which are generally grown on less than 300,000 acres per crop. ARS participates in a State-Federal program known as IR-4 to assist in the development of data to support pesticide residue tolerances established by the EPA, used by the pesticide registrants to add the crops as approved uses. In 2010, ARS scientists established 137 pesticide/crop combinations at field locations in seven States (South Carolina, Arizona, California, Georgia, Washington, Texas, and Ohio) to treat with pesticides. In laboratories in Beltsville, Maryland; Wapato, Washington; and Tifton, Georgia, ARS scientists analyzed 163 pesticide/crop combinations for pesticide residue tolerances. ARS contributed data for pesticide residue tolerances on 26 specialty crops and 15 pesticides that can be used by registrants to label these as available to specialty crop growers. (NP 304; C2, PS 2B; PM 4.2.4; Project # 0500-00007-089-00D)

Ecological clues to stink bug control in the South. Three major stink bugs (southern green, brown, and green) affect key southern crops such as cotton, corn, and peanut; which are often grown in rotation. Ecological and behavioral studies by ARS researchers in Tifton, Georgia, have shown that natural enemies of stink bugs, including tiny parasitic flies, predatory fire ants, spiders, and others are responsible for significant control, particularly when border vegetation is conserved, and that early planted corn suffers less stink bug damage than late planted corn. Overall results suggest that there are crop-specific predator species that are able to cause high mortality of stink bug egg masses. This work will be employed by those combating the newly invasive brown marmorated stink bug. In addition, the researchers demonstrated for the first time that the southern green stink bug can be trapped with its pheromone. The results suggest that stink bugs trapped with a pheromone blend from all three stink bugs has the greatest potential for detecting all of the stink bugs in diversified agro-ecosystems. (NP304; C2, PS 2A&2B; PM 4.2.4; Project # 6602-22000-036-00D)

Preceding crop affects soybean tolerance to weed pressure. There is an urgent need for effective cultural tactics that reduce emerging weeds in crop rotations that include corn, soybean, and wheat. ARS researchers in Brookings, South Dakota, found that tolerance of soybean to weed infestation doubled when following corn than when following spring wheat, oat, dry pea, or soybean. The research demonstrated that crop diversity, combined with the specific sequence of the crop species in the rotation design, disrupts normal growth of weeds and reduces weed community density over time. Cropping sequences that increase tolerance to weed interference also reduce the impact of weeds on crop yield. These sequences will be especially helpful to soybean and corn growers seeking to reduce herbicide use while accepting a low density of weeds without harming the crop. Producers using the strategies developed in this research have reduced herbicide use by 30 to 50 percent compared with previously

recommended management. (NP304; C2, PS 2B; PM 4.2.3; Project # 5447-21220-003-00D)

New grass demography data improves re-vegetation success in rangeland restoration. Degraded rangeland can be difficult to restore because invasive species can often be successful in these systems, while establishment of native species' seedlings is not. Augmentative seeding can help overcome this limitation, however it can be expensive and success rates are variable. ARS scientists in Burns, Oregon, examined the demography of grass species to determine at what life stage seeding failure is most likely, and determined that the most critical period for native species establishment was the transition between germination and emergence. They also demonstrated that important plant traits for establishment for both invasive and native species in low nitrogen soils, such as degraded rangeland, were early germination, root growth at low temperature, and a high specific leaf area. Also, ARS scientists at the Pest Management Research Unit, in Sidney, Montana, demonstrated that the environment in which plants are grown can impact the traits carried over to seed progeny, i.e., drought-stressed plants produce drought-tolerant seeds. The results of these studies will be of value to researchers and land managers in the selection and use of native species lines in the restoration of degraded rangeland. (NP304; C3, PS 3B; PM 4.2.4; Project # 5360-22000-003-00D & 5436-22000-013-00D)

Chemical pesticides for growers of nursery and floral crops. The Ornamental Horticulture Program supports an industry valued at over \$16.9 billion in annual sales and crops which are grown under a number of conditions such as nurseries, greenhouses, and tree farms. The plants can be in beds, containers or in-ground. The growers are involved in a number of diverse markets including flowers, bulbs, houseplants, perennials, trees, shrubs, nonbearing fruit trees, and others. Treating such an enormous variety of plants with pesticides presents a challenge for crop safety, so considerable that effort must be spent in developing phytotoxicity data so that pesticide manufacturers will add these crops to their labels. In 2010, ARS scientists established 210 pesticide/crop combinations at field locations in six states (South Carolina, Mississippi, Georgia, Washington, Texas, and Ohio) to treat with pesticides and evaluate them for crop safety. A select number of these combinations were also evaluated to see how well the pesticide performed against the target pest. ARS contributed data toward the registration of uses for 112 crops and 23 pesticides that are now available to growers of florist and nursery crops to reduce losses from pests. (NP304; C2, PS 2B; PM 4.2.4; Project # 0500-00007-089-00D)

Control strategy mitigates the threat of the invasive Argentine cactus moth in the United States and eradicates the pest in Mexico. Since its detection in south Florida in 1989, the Argentine cactus moth has expanded its range each year along the Atlantic Coast and west along the Gulf Coast to the barrier islands of Mississippi. This moth has become an imminent threat to many *Opuntia* cactus species which are valued as food, forage, wildlife habitat, and a major plant group contributing to ecosystem structure and biodiversity. ARS researchers in Tifton, Georgia and Tallahassee, Florida, in collaboration with the USDA Animal and Plant Health Inspection Service (APHIS), developed and refined survey methods and control tactics, using field sanitation combined with sterile insect releases, along the leading edge of the invasion and at new outbreak locations. With the cooperation of ARS, APHIS, and SAGARPA (Mexico's department of agriculture), Mexico continues to adopt and implement these methods and tactics in the operational program that is part of an ongoing United States-Mexico bi-national campaign against this invasive pest. These actions have eradicated or greatly reduced established populations of this pest on the Alabama and Mississippi barrier islands and the islands off the coast of Quintana Roo, Mexico, as well as mitigated the further westward expansion of pest populations along the Gulf coast. This is the first time any moth pest has been eradicated from a country in the Western Hemisphere. (NP304; C2&3, PS 2A, 2B & 3A; PM 4.2.4; Project # 6602-22000-035-00D)

A novel approach for detecting Russian wheat aphid infestations in wheat fields. The ability to quickly categorize pest status in large wheat fields is critical to facilitating timely application of control measures, but there are no efficient methods available to sample large fields at this time. ARS researchers in

Stillwater, Oklahoma, with collaborators, developed remote sensing technology to detect and monitor infestations of Russian wheat aphids in production winter wheat fields. Over the duration of this project, airborne multispectral imagery was acquired from numerous production wheat fields in western Oklahoma and southeastern Colorado, then processed and analyzed using both standard and novel analytical methods. Stress caused by the Russian wheat aphid could be detected with multi-spectral imagery of infested wheat fields. Using a combination of spectral information combined with spatial images of stressed plants, stress caused by Russian wheat aphid could be differentiated from non-stressed fields and fields that were stressed by common environmental factors such as drought. In the future, the ability to categorize the pest status for fields using airborne multi-spectral remote sensing will facilitate timely application of control measures without the need for expensive and time-consuming within-field pest scouting. (NP304; C2, PS 2A; PM 4.2.3; Project # 6217-22000-014-00D)

New biocontrol agents identified for Cape Ivy. Cape ivy is an invasive alien weed from South Africa that smothers native vegetation along the coast of California. ARS scientists in Reno, Nevada, conducted laboratory experiments to evaluate the host (Cape ivy) specificity of a stem-boring moth and a gall-forming fly. Both species are highly specific to Cape ivy, and a petition demonstrating their safety for release was submitted to the USDA APHIS Technical Advisory Group for Biological Control of Weeds. Successful establishment of these agents should reduce the size and abundance of the Cape Ivy weed, reduce control costs, and lead to the reestablishment and improved survival of native vegetation. (NP304; C3, PS 3B; PM 4.2.4; Project # 5325-22000-020-00D)

Weed-free crop rotation reduces Corky Ringspot disease. Corky ringspot disease (CRS) of potato is caused by tobacco rattle virus (TRV) and vectored by stubby root nematode and makes tubers unmarketable. CRS is present in approximately 5% of the potato acreage of the Columbia Basin and soil fumigation, costing \$200/acre, is currently the only method of control. Researchers at the Vegetable and Forage Crops unit in Prosser, WA demonstrated that growing weed-free alfalfa helps prevent CRS. Weeds that were identified as hosts of TRV and stubby root nematode and present in the crop rotation prevented the elimination of TRV from the nematode population. By using alfalfa in the rotation and keeping it free of these weeds, growers can lessen the need for costly soil fumigation, saving approximately \$1.5 million dollars to the industry. (NP304 C2 PSB; PM 2.2.1 #5354-21660-002-00D)

Human Nutrition (ARS Goal 5) (107)

Select Examples of Recent Progress:

NP107

Adequate vitamin D reduces the risk of falls in the elderly. Falls are a major cause of fractures and other injuries in the elderly. Vitamin D deficiency was determined to be a major risk factor for falling. Conversely, supplementing the diet with vitamin D and correcting vitamin D deficiency, combined with moderate home-based exercise, significantly decreased the risk of falling. Scientists at the ARS laboratory in Boston, Massachusetts conducted a meta-analysis on all available data and concluded that at least 700 international units of vitamin D was needed daily. These data were recently used by the National Academy of Science's Institute of Medicine to increase the recommended amount of vitamin D for people over the age of 70. (NP 107, C 2, P.S. 2A, P.M. 5.2.2, Project # 1950-51000-069-00D)

Moderate exercise alone does not increase overall energy expenditure in teens. In the debate over the relative roles of diet and exercise in controlling weight, it is unclear what effect each has on maintaining healthy body weight. Researchers at the ARS laboratory in Houston, Texas found that 12 weeks of moderate intensity exercise for 30 minutes four times per week in lean and obese sedentary adolescents did not change overall energy expenditure. Fat oxidation increased in the lean subjects. However, fat

accumulation in the liver and abdomen decreased in obese adolescents. The data suggests that even more exercise and/or calorie restriction is an essential component of a successful weight loss strategy and that obese teens require more exercise than their lean counterparts to increase fat burning. (NP 107, C 3, P.S. 3A, P.M. 5.2.2, Project #6250-51000-053-00D)

A variation in a common gene is associated with food intake and obesity. The epidemic of obesity and overweight has resulted in a critical need for methods of predicting risk to an individual as well as predicting the possible benefit of therapeutic interventions. ARS funded researchers in Boston, Massachusetts conducted a large study to investigate the role of variation in a specific gene (APOA2 - 265T) for regulation of food intake and body weight. People carrying this genetic variation only developed obesity when they consumed a diet high in saturated fat. This finding will contribute to efforts to identify individuals susceptible to diet induced obesity, and will ultimately allow for specifically tailored dietary recommendations to reduce their risk of obesity and cardiovascular diseases. (NP 107, C 3, P.S. 3A, P.M.5.2.2, Project #1950-51520-012-00D)

Ratio of animal to vegetable protein has little effect on heart disease risk factors. Researchers from the ARS lab in Boston, Massachusetts tested the long-standing hypothesis that the ratio of two amino acids, lysine and arginine, that differ considerably in animal and vegetable proteins, affects serum cholesterol and other risk factors for heart disease. Feeding 30 adults controlled diets resulted in only a few small differences out of more than two dozen risk factors tested, including no effect on any of the standard serum cholesterol measures. This theory has been studied for close to 40 years and this experiment is likely to end the debate; it also suggests that animal protein does not increase risk for heart disease more than vegetable protein. (NP 107, C 2, P.S. 2A, P.M. 5.2.2, Project #1950-51000-072-00D)

New national dietary intake results, data briefs, and database released. ARS scientists at Beltsville, Maryland released nationally representative information for 2007-2008 from the "What We Eat in America" diet survey portion of NHANES for use by other researchers. In addition, 36 data tables summarizing those results and data briefs on milk consumption and adolescent snacking provide useful information to researchers, clinicians, and policymakers. A new version of the "USDA Food and Nutrient Database for Dietary Studies (FNDDS)," along with a search tool to use FNDDS, were also made available to other government agencies that enable them to calculate nutrient intakes from food consumption data. All of these products provide the most accurate and current knowledge of food and nutrient intake in the U.S. (NP 107, C 1, P.S. 1A, P.M. 5.2.1, Project #1235-53000-016-00D)

Controlling stress may result in selection of a better diet. The roles of stress or stress hormones and their relationship to food choices and body weight/body composition of middle aged women are unknown. ARS researchers at Davis, California found that greater perceived stress was associated with lower fruit, vegetable, and protein intake; greater consumption of salty snacks; lower participation in physical activity; and a higher intake of sweets, particularly in those with type 2 diabetes. Many of these effects were related to concentrations of the hormone cortisol. These findings demonstrate that stress reactivity influences food choice and food intake, and that controlling stress may have a large impact on factors associated with obesity. (NP 107, C 3, P.S. 3B, P.M. 5.2.2, Project # 5306-51530-019-00D)

Inclusion of whole grains and/or fiber-rich cereals in the diet correlate with less abdominal fat deposition. Foods high in dietary fiber may play an important role in regulating body weight but few studies have examined the relationship between dietary fiber from different sources and body fat. ARS funded researchers at Boston, Massachusetts, examined the relationship between grain intake and measures of body fat. Higher whole-grain and cereal fiber intakes were associated with improved body mass index, less total body fat and less fat in the abdominal region (the fat mass that is most strongly associated with obesity-linked disorders). This research suggests that increasing intakes of cereal fiber, particularly from

whole-grain sources, may be one means of limiting body fat and decreasing the risk of obesity-related health disorders. (N.P. 107, C 3, P.S. 3A, P.M. 5.2.2, Project # 1950-51530-009-00D)

<u>Maternal obesity programs energy balance in offspring.</u> Researchers in Little Rock, Arkansas found that male offspring of obese rats gain more weight and fat mass, along with developing insulin resistance, when fed high-fat diets. The scientists identified multiple genes and proteins in the liver and blood that change before development of obesity, suggesting that *in utero* exposure to maternal obesity programs multiple aspects of energy balance and offers potential ways to control weight gain if the same metabolic changes occur in humans. (NP 107, C 4, P.S., P.M. 5.2.2, Project #6251-51000-007-00D)

New, inexpensive method to detect variation in nutritional content of food. The chemical composition of plants varied significantly as a function of genetics and the environment (weather, soil, and other biota) according to ARS scientists in Beltsville, Maryland who combined ultraviolet spectroscopy of bean extracts with pattern recognition computer analysis to demonstrate differences in the composition of 9 cultivars in 3 growing locations (MD, MI, and NE). In addition, the plant-to-plant variation was determined to be approximately 70%. This research demonstrated that an inexpensive analytical method can be used to detect subtle differences in chemistry arising from micro- (plant-to-plant) and macro- (different states in the US) environments and genetics (cultivar). The low cost of the method makes it an easily accessible tool for all plant researchers. (NP 107, C 1, P.S. 1C, P.M. 5.2.3, Project #1235-52000-060-00D)

Development of a standard reference material for infant and adult nutritional formula. Standard Reference Materials are substances that are used as the "Gold Standard" when unknown foods or substances are being analyzed. Standard Reference Materials are essential for the development of Quality Control programs and for conducting analyses that are the basis for food labeling and regulations. ARS chemists at Beltsville, Maryland, worked with the National Institute of Standards and Technology to develop and release a standard reference material for "Infant/Adult Nutritional Formula" (SRM® 1849). This material is available nationally and internationally for use by all laboratories that wish to analyze unknown nutritional formula, and use of this Standard Reference Material will facilitate the improvement and enforcement of quality controls on the manufacture of nutritional formula. (NP 107, C 1, P.S. 1C, P.M. 5.2.3, Project # 1235-52000-059-00D)

Environmental Stewardship (ARS Goal 6) (211, 202, 212, 206, 215, 216, 308)

Select Examples of Recent Progress:

Management Initiative 2: Ensure Provision and Permanent Access of Quality Agricultural Information for USDA, the Nation, and the Global Agricultural Community via the National Agricultural Library

Select Examples of Recent Progress:

NP211

Satellite-derived Evaporative Stress Index reproduces spatial and temporal variability in rainfall patterns without monitoring precipitation. Compared to traditional weather and precipitation monitoring networks, remotely-sensed data collected by satellites have the potential to more readily and accurately assess and predict drought. In particular, thermal infrared (TIR) remote sensing data carry valuable information about surface moisture availability. ARS scientists at Beltsville, MD used Geostationary

Operational Environmental Satellite (GOES) TIR imagery and a fully automated inverse model of Atmosphere-Land Exchange to produce hourly evapotranspiration (ET) estimates for a 10-km resolution grid covering the contiguous United States, and then used these estimates to develop and evaluate an Evaporative Stress Index derived from temporal anomalies in the ratio of actual-to-potential ET. Compared to standard drought metrics, this remotely sensed ET index captures spatiotemporal variability in rainfall patterns without the need for precipitation data, and could thus be particularly useful in data-sparse parts of the globe where accurate drought prediction could help to improve food and water security. The model has been revised to use standard National Oceanic and Atmospheric Administration (NOAA) satellite and meteorological data, with operational processing is being transitioned to NOAA for incorporation into the Climate Prediction Center's North American Drought Briefing. Products are also being evaluated by the National Drought Mitigation Center. (NP211 C1 PS- E; PM 6.1.1; #1265-13610-027-00D)

Guidelines and protocols developed for regional-scale salinity assessment of the Red River Valley. Land managers and policy makers across the globe need a regional-scale tool for measuring and inventorying soil salinity in agricultural fields where salt buildup lowers crop yields. A scientific team led by an ARS scientist at Riverside, CA, used MODIS imagery to assess and map soil salinity across 300,000 hectares of North Dakota's and Minnesota's Red River Valley (RRV). Rising soil salinity levels due to rising water tables in the RRV have been linked to increasing precipitation resulting from climate change. They found that 53 percent of the variability in soil salinity could be correlated to multi-year enhanced vegetative index and whether land was eligible for Conservation Reserve Program inclusion (a federal program that sets aside marginally productive land based on conservation principles). Results from this research provide the NRCS with protocols and guidelines for mapping soil salinity over hundreds of thousands of hectares of the RRV and has applicability for many other regions of the world. (NP211 C1 PS-B; PM 6.1.1; #5310-61000-013-00D)

Climate change affects intermountain hydrology. A variety of studies have demonstrated links between increasing temperatures, declining snowpacks, and earlier streamflow in snow fed streams in the western U.S., but in all of these studies the linkages are based on observations collected from multiple locations. ARS scientists at the Northwest Watershed Research Center in Boise, Idaho analyzed 45 years of air temperature, snow, precipitation, and streamflow data measured at the same location, the Reynolds Creek Experimental Watershed. Over the period of record, average air temperature has increased about two degrees centigrade, snow disappears more than a month earlier at lower elevations, and while total annual precipitation has remained constant, the onset of streamflow now occurs earlier in the season. Because they demonstrate the inadequacy of current management approaches, these changes have major implications for reservoir management in the West where the vast majority of agriculture is irrigated. (NP211 C1 PS-E; PM 6.1.1; #5362-13610-008-00D)

Weather and crop water use network provides updated regional water use projections for next 50 years in northern Texas. In the northern Texas High Plains, where irrigation accounts for 90% of all water use, accurate and representative irrigation water use estimates are necessary for sustainable production agriculture. ARS and Texas AgriLife Research updated and improved evapotranspiration (ET) data from the Texas High Plains ET Network (TXHPET). The data are used to compute water use and irrigation water demand estimates for all crops grown in the Texas Panhandle. The ET data are used with Farm Service Agency crop acreage data and precipitation on a county basis to run the Texas A&M—Amarillo (TAMA) irrigation demand model, which was modified to include new crop categories and yearly and forecasted ET data inputs. Use of the accurate and representative TXHPET network-based water use data reflects regional production potential as well as actual reductions in water use by producers using new and improved irrigation management, technology and methods. This information is being used by regional water planners for water demand planning purposes, by groundwater water conservation districts in

establishing pumping regulations, and for regional socio-economic and sustainability analyses. (NP211 C1 PS-B, C1 PS-E; PM 6.1.1; #6209-13000-012-00D)

Sediment and nutrient benefits of Conservation Reserve Program land use conversion quantified. Billions of dollars have been invested to remove cultivated lands from production for conservation purposes via the Conservation Reserve Program (CRP), but the resulting environmental benefits have rarely been quantified. ARS scientists in Oxford, MS, showed that in the Mississippi Delta, converting 280 ha of cropped land to trees reduced sediment concentrations and nutrient loads in runoff by an order of magnitude relative to adjacent sites with similar topography and soils that were under reduced tillage crop production. Associated positive impacts on the water quality of the receiving water body, Beasley Lake, have also been measured. These findings support future federal investment in the CRP. (NP211 C1 PS-A; PM 6.1.1, #6408-13660-006-00D)

Release of updated WEPP model version 2010.1 in January 2010. WEPP model version 2010.1 contains significant improvements in the prediction of soil freezing and thawing, as well as snow accumulation and runoff resulting from snow melting. Cooperative work between ARS-NSERL, ARS-Pullman, and Washington State University resulted in this updated version. The new model version performs much better in validation studies using data from Pullman, WA and Morris, MN. Additionally, some other important corrections were made to the code that fixed problems related to soil layer representation and multiple flow element erosion simulation. This model release impacts the thousands of WEPP model users throughout the U.S. and world, and will provide better predictions of runoff, soil loss and sediment yield from hillslope profiles and small field-scale watersheds. Users in areas experiencing substantial erosion from snowmelt on thawing soils should notice particularly better performance. (NP211 C1 PS-D; PM 6.1.1; #3602-12000-012-00D)

Potential of drainage water management to improve water quality in the midwestern U.S. quantified. Drainage water management (DWM) is a promising technology for reducing nitrate losses from artificially drained or "tiled" fields. While there is an extensive history for the practice in North Carolina, little is known about the efficacy or cost effectiveness of the practice under Midwest conditions where artificial drainage is widely used. ARS scientists at Ames, Iowa used soil and land cover databases combined with modeling to estimate that 4.8 million hectares of land currently used to grow corn in the Midwest would be suitable for DWM, with the potential to reduce nitrate loss by approximately 83,000 metric tons (91,300 tons) per year. Considering the cost of control structures, redesign of new drainage systems, and payments to farmers to adjust the control structures to reduce nitrate losses, the cost per kilogram of nitrate reduced in drainage water for DWM was estimated at \$2.71 (\$1.23/lb). This information will be useful to farmers and State and Federal action agencies in setting priorities for the expenditure of conservation monies to improve surface water quality. (NP211 C1 PS-E; PM 6.1.1; #3625-13000-009-00D)

Closing the curtain on phosphorus losses from agricultural fields to drainage ditches. The loss of phosphorus in shallow groundwater leaving fields of the Delmarva Peninsula can account for more than 90% of the phosphorus exported by drainage ditches to tributaries of the Chesapeake Bay. ARS scientists at University Park, PA, installed a curtain of gypsum along the border of a field from which high loads of phosphorus had previously been recorded in groundwater. The new curtain, or permeable reactive barrier, removed 38-59% of the dissolved phosphorus in groundwater in the first rain storms after it was installed. Efforts are underway to expand the testing of this water quality enhancement practice in other areas of the region. (NP211 C1 PS-E; PM 6.1.1; #1902-13000-011-00D)

Aerial photography used for mapping giant reed infestations along the Texas-Mexico portion of the Rio Grande. Giant reed is an invasive weed throughout the southern half of the United States, with the densest stands growing along the coastal rivers of southern California and the Rio Grande in Texas. ARS

scientists at Weslaco, TX, used aerial photography to map giant reed infestations and estimate infested areas along the Texas-Mexico portion of the Rio Grande. Based on the aerial photographs and ground surveys, the portion of the river from San Ygnacio to Lajitas that has a river length of 898 km was found to be infested with giant reed, with the densest populations located between Laredo and Del Rio. The total giant reed area along the Rio Grande between Lajitas and San Ygnacio was estimated to be 5985 ha, with 3670 ha or 61% on the U.S. side and 2315 or 39% on the Mexican side. This study provides not only the methods for quantifying giant reed, but also the first accurate estimates of giant reed infestations along the Texas-Mexico portion of the Rio Grande. This information was necessary to gain approval for release of biological control agents from government agencies in the U.S., Mexico, and Canada. The study will contribute to estimates of water usage of giant reed, and will also be useful for both land owners and government agencies for the management and control of giant reed. (NP211 C1 PS-E; PM 6.1.1; #6204-11660-007-00D)

Design and management criteria for fish, amphibian, and reptiles within created wetlands. The ability of created wetlands to reduce nutrient, pesticide, and sediment loadings within agricultural runoff can be optimized by design and management; These priorities, however, may result in wetlands that are not beneficial as habitats for aquatic vertebrates such as fishes, amphibians, and reptiles, which are exhibiting worldwide population declines. ARS scientists in Columbus, OH documented differences in fishes, amphibians, and reptiles between two wetland types created as a result of the wetland-reservoirsubirrigation system (WRSIS) and used the information to develop design and management criteria capable of increasing the ecological benefits resulting from this agricultural water recycling system. Differences in amphibian abundance and species composition between WRSIS wetlands and reservoirs suggest the potential for WRSIS wetlands to provide habitat for a different community of aquatic vertebrates than the reservoirs. Furthermore, WRSIS dominated by fishes did not exhibit the benefits of a two stage wetland design. A set of design and management criteria were developed that will enable WRSIS wetlands to be managed as amphibian habitat and reservoirs to be managed as fish habitat. These design and management criteria can also be used by state, federal, and private agencies involved with creating agricultural wetlands to assist them meeting their conservation and restoration goals. (NP211 C1 PS-F; PM 6.1.1; #3604-13000-008-00D)

NP202

Physical/chemical fractionation procedures developed for SOM characterization. The complex nature of soil organic matter (SOM) was more completely characterized by using innovative physical/chemical fractionation procedures in combination with state-of-the-art spectroscopic techniques. ARS scientists in Ames Iowa devised an innovative, integrated SOM fractionation/characterization procedure that permitted sequential isolation of SOM fractions obtained using physical or chemical separation of the same soil sample. Multiple laboratory analyses were used to characterize and compare elemental and chemical composition of several physically- or chemically-defined SOM fractions with whole SOM. This method is being used to detect subtle differences in carbon pools and nutrient cycling due to soil and crop management, such as tillage, crop rotation, cover crops, and bioenergy feedstock production. (NP 202, C 3, P.S. 1, P.M. 6.2.1, #3625-11120-003-00D)

Cost of nutrient losses in furrow irrigation runoff. Nutrients in furrow irrigation runoff are a lost resource. ARS researchers at Kimberly, Idaho, measured the amount of phosphorus, inorganic nitrogen, and potassium lost in runoff from furrow irrigated fields which were fertilized conventionally or with manure. Most of the nutrient loss was phosphorus, and most of the phosphorus loss was associated with sediment. Less than 3% of the applied nitrogen was lost (2.7% of total inorganic N applied to fertilizer plots, 1.5% of total N applied on manure plots). About one third of the phosphorus applied with the manure was lost with runoff during the two year study (84 lb P/acre lost out of 219 lb P/acre applied in two years). No

phosphorus was applied to the fertilizer or control treatments, which had losses of 33 to 48 lb P/acre /year. The cost to replace nitrate, ammonia, potassium and phosphorus from control plots averaged \$35/acre/year; and \$40/acre/year for fertilizer and manure treatments. This research underscores the need for management practices to reduce nutrient losses from furrow-irrigated soils. (NP 202, C 4, P.S. 1, P.M. 6.2.1, #5368-12000-009-00D)

<u>Herbicides affect nutrient cycling.</u> Farmers have observed symptoms of nutrient deficiency in crops where the herbicide glyphosate has been used for several years in a row. ARS scientists in West Lafayette, IN found that one reason for this is that long term exposure to glyphosate affects soil enzyme activities that are critical in nutrient cycling. These findings will impact fertilizer application rate and weed management decision making with soil quality and crop yield in mind. (NP 202, C 1, P.S. 1, P.M. 6.2.1; NP 202, C 7, P.S. 1; P.M. 6.2.1, #3602-12220-006-00D)

Biochar can be used to increase carbon storage and improve soil quality in Midwestern fields. Biochar is a form of charcoal produced while making renewable biofuels through the process of pyrolysis. Agricultural Research Service (ARS) scientists in Ames, IA, conducted laboratory and field studies to determine how biochar affected several soil biological, chemical, and physical properties in a typical agricultural soil. The laboratory study showed that applying biochar decreased soil compaction, increased soil acidification, and increased the capacity of soil to hold water and plant nutrients compared to soils that did not receive biochar. Applying biochar increased total nitrogen by as much as 7%, organic carbon (C) by as much as up to 69%, and the quantity of several other plant nutrients compared to the unamended soil. These results are important because pyrolysis produces a co-product that can be used to increase soil carbon storage and improve other soil properties and processes (i.e., soil quality). (NP 202, C 3, P.S. 3, P.M. 6.2.1, #3625-11120-003-00D)

No evidence found that Bt corn residues decompose more slowly than non-Bt residues. ARS scientists at Brookings, South Dakota have completed a series of studies published in three papers that found no evidence that the decomposition of corn residues was linked to the presence of the Bt gene in the corn hybrid. These studies examined multiple corn hybrids from a single manufacturer, hybrids from different seed manufacturers, and hybrids grown under conditions of differential insect pressure. With the rapid expansion of genetically modified crops, controlled studies evaluating the potential for unintended effects due to genetic modification provide a firm basis for evaluating the advantages and disadvantages of this technology. (NP 202, C1, P.S. 1, P.M. 6.2.1, #5447-12620-002-00D)

Corn yield increased by applying fertilizer with strip tillage. Strip tillage is a conservation practice that can incorporate fertilizer into the soil while leaving much of the surface undisturbed. This tillage practice can potentially increase corn yield by applying fertilizer directly beneath the corn row. ARS researchers at Kimberly, Idaho determined that applying nitrogen and phosphorus fertilizer with strip tillage increased corn yield on eroded soils 12 percent compared to broadcast fertilizer application, and 26 percent compared to surface banding with the planter. Reduced tillage costs and potential increased corn yield with strip tillage could increase the economic productivity of eroded land in the Pacific Northwest. (NP 202, C 4, P.S. 2, P.M. 6.2.1, #5368-12000-009-00D)

<u>Fungicide use substantially slows degradation of a widely used weed control product.</u> Pesticides are a diverse group of chemicals including products used to control weeds (herbicides), plant diseases (fungicides), and insects (insecticides). During large scale production of most crops, pesticides from one or more of these classes are applied to farm fields. A common scenario in peanut production in the Southeastern USA is herbicide application at planting followed by fungicide applications after the crop emerges. Fungicides may alter and impact soil microbial communities thus there is a potential to impact herbicide degradation, and change herbicide efficacy and environmental fate characteristics. ARS researchers at the Southeast Watershed Research Laboratory in Tifton, Georgia evaluated potential

impacts of four fungicides widely used by peanut growers on the soil persistence of a commonly used herbicide (metolochlor). One of the fungicides was found to increase persistence of the herbicide in soil by more than 2 times. This information is of considerable value to peanut growers and other farmers in the region who use these chemicals. Modified farming practices, such as reducing herbicide application rates, may need to be considered where the fungicide and herbicide are used in combination. (NP 202, C 1, P.S. 1; C 3, P.S. 7; P.M. 6.2.1, #6602-12130-001-00D)

Speeding up soil quality recovery in sandy soils under dryland production. Since the 1940s much of the land in the Southern High Plains region has been planted solely to cotton using conventional tillage practices, resulting in declining soil quality. In order to sustain future agricultural activities in this area, conservation practices such as minimum tillage and cotton rotations with sorghum are being implemented to reduce erosion and restore soil quality in dryland production. But it often takes many years before improvements can be seen. ARS scientists at Lubbock, TX found that introducing a rotation of cotton with high biomass crops, such as forage sorghum and a winter rye cover crop, produced increases in microbial biomass carbon and nitrogen, and the activities of enzymes important for nutrient cycling in as little as three years. It took over five years to show improvements under rotations of cotton and grain sorghum compared to cotton monoculture. This is new information that growers and researchers can consider when identifying cropping systems that will result in improved soil quality and functioning. (NP 202, C 2, P.S. 1; C 3, P.S. 3; C 5, P.S. 1; P.M. 6.2.1, #6208-12000-009-00D)

Improved productivity of sandy coastal soils. Because the southeastern U.S. Coastal Plain is hot and wet, its weathered sandy soils have low organic carbon (C) contents and poor productivity. A study was conducted to test the potential benefits of soil additions of biochar, a byproduct of bioenergy production by pyrolysis of biomass. ARS researchers at Florence, SC, added C to a coastal soil in the form of non-activated pecan biochar and, as a comparison with a more conventional carbon source, finely ground switchgrass. Both biochar and switchgrass additions decreased soil compaction. Switchgrass additions, however, also increased the water holding capacity of the soil while the biochar additions did not. These improvements enhance the quality of these degraded and weathered sandy soils and sequester carbon. (NP 202, C 9, P.S. 1, P.M. 6.2.1, #6657-12000-005-00D)

Grassland management for increased biomass production and soil carbon (C) storage. Producers considering biomass production or soil C storage using native grass species need scientifically-based information on management options. ARS scientists at Brookings South Dakota, in collaboration with scientists at South Dakota State University, are conducting field research which revealed that methods used to manage grass canopies had significant effects on the rates of biomass production and soil C accumulation during the first 9 years of the long term experiment. If the producer's goal is to maximize harvested grass biomass for sale as a feedstock, an annual late-summer haying treatment could be recommended with the understanding that this would be slightly less efficient at increasing the soil C accumulation rate. A spring burn treatment would be recommended if the goals were to restore or maintain dominance of warm season grasses in mixed grasslands while only slowly increasing soil C accumulation, while no canopy management would be recommended if the primary goal was to increase soil C accumulation. This customer-driven research provides biomass and soil data that will aid farmer decisions related to the type of grassland management strategies that best fit their current and future agronomic needs. (NP 202, C 5, P.S. 1, P.M. 6.2.1, #5447-12620-002-00D)

NP212

<u>New model for estimating ammonia emissions from dairy manure.</u> There is a need to update tools for predicting ammonia emissions from dairy livestock manure as existing models are based upon old data from obsolete dairy facilities. ARS scientists at University Park, PA collected data from a range of

modern dairy facilities and developed new equations that were found to better estimate ammonia emissions from dairies currently being operated. The new findings are being integrated into a decision support tool that will enable producers to evaluate the effects of nitrogen management and thus ammonia mitigation strategies, on the total air emissions from farms. Use of the updated decision support tool enables increased nitrogen use efficiency, which in turn reduces ammonia emissions and can increase profit margins. Reducing ammonia emissions improves both air, and water quality via reduced deposition of atmospheric nitrogen. (NP 212 C2 PS-B; PM 6.2.1; #1902-11130-001-00D)

Simplified method for determining the susceptibility of soils to wind erosion. The wind erodible fraction (WEF) is a measure of the susceptibility of soils to erosion by wind and provides a useful tool for planning farming production strategies. The standard method for measuring WEF uses a laboratory-based rotary sieve that requires transportation of soil samples to the laboratory. Unfortunately, soils tend to break up during transport to the laboratory, yielding erroneous results. A rapid and simple field-based sieving protocol for determining WEF was developed by ARS researchers in Manhattan, Kansas. The protocol was described in a report delivered to Natural Resources Conservation Service (NRCS) and is now being used by NRCS in the field on a limited test-basis as a quick method for determining temporal WEF. This method is also being incorporated into the NRCS National Agronomy Manual of policy and procedures for wind erosion. NRCS is now better-equipped to help landowners increase air quality and improve soil sustainability via reductions of wind-eroded soil. (NP212 C1 PS-B; PM 6.2.1; #5430-11120-008-00D)

New method to "fingerprint" particulate matter emitted from cattle feedlots. Current methods to evaluate particle concentrations from cattle feedlots fail to identify the sources, thus limiting progress towards strategies to reduce particulate emissions. Using Raman microscopy methods, ARS scientists from Beltsville, Maryland examined the chemical profile of individual particles captured downwind from a cattle feedlot and compared them with the chemical profiles of particles taken from potential sources within the feedlot. Positive matches enabled sourcing of airborne samples to a specific location within the field lot. This approach can be used to determine the most important sources of particles to the total particle emission flux. Detailed information on particle sources will be useful for evaluating the efficacy of existing and future improved dust management practices. (NP212 C1 PS-A, C1 PS-B; PM 6.2.1; #1265-12610-001-00D)

An improved remote sensing method for assessing crop residue cover. Crop residue cover, a product of low-till and no-till farming, is an important tool for controlling soil erosion, enhancing soil productivity and increasing soil carbon sequestration. Current methods of measuring residue cover for management decisions are inadequate for characterizing the spatial variability of residue cover over many fields. Measurements based on Landsat satellite data for mapping residue cover are often confounded by changes in how the soil itself reflects light. There are reliable ways to get the data based on very narrow ranges of reflected wavelengths, but that kind of imagery is expensive and not widely available. An alternative approach was proposed and tested at five locations in the USA on multiple dates. Results show that a minimal upgrade from one broad Landsat wavelength band to two or three relatively narrow bands would provide reliable estimates of crop residue cover and soil tillage intensity data for regional assessments of conservation practices. This work advances crop residue mapping, as well as biogeochemical models that produce credible estimates of soil carbon at watershed and regional scales. (NP212 C1 PS-A; PM 6.2.1; #1265-12130-002-00D)

Rising atmospheric CO2 favors weedy rice over cultivated rice. Rising atmospheric CO2 increases the growth of many crops and enhances the growth of weeds. Weedy red rice is a troublesome weed in cultivated rice fields that reduces the quantity and quality of rice harvested for food. Research showed that weedy red rice growth increased more with additional CO2 than cultivated rice. Further, higher CO2 increased the ability of weedy red rice to compete with cultivated rice to the detriment of rice yield.

These results document greater susceptibility of rice production to in-field competition from greater weed vigor driven by rising atmospheric CO2, and highlight the need to assess the combined impacts of increasing atmospheric CO2 and rising air temperatures on global rice production.

(NP212 C3 PS-A; C3 PS-C; PM 6.2.1; #1265-11210-001-00D)

Predicting plant invasions in an era of global change. Global change is expected to dramatically alter invasive plant occurrences, and will require corresponding changes of land management. ARS scientists from Cheyenne, WY and Ft. Collins, CO, collaborated with two Princeton University scientists, to provide a new framework for examining this issue in an invited review and synthesis article. Supported by findings from both modeling and experimental studies, the scientists concluded that global changes that increase the availability of plant resources, such as atmospheric carbon dioxide (CO2) enrichment and nitrogen (N) pollution, are most likely to cause plant invasion. They proposed management changes needed to combat invasive plants in the face of global change. This work will be used extensively, both by global change biology researchers to better understand climate change impacts on ecosystems, and by managers of natural and semi-natural ecosystems. (NP212 C3 PS-B; PM 6.2.1; #5409-11000-004-00D)

Influence of nitrogen fertilizer forms on greenhouse gas emissions from corn depends on production and environmental conditions. A significant source of nitrous oxide (N2O), a greenhouse gas with significantly greater impact on climate change than carbon dioxide emissions, is from the use of nitrogen (N) fertilizers. Use of enhanced-efficiency N fertilizers (controlled release and stabilized nitrogen sources) was shown to influence the amount of N2O emitted from irrigated no-till corn. In no-till irrigated corn in the semiarid western U.S, nitrous oxide emissions monitored by ARS scientists in Fort Collins, Colorado during the 2007 and 2008 growing seasons were reduced by the use of enhancedefficiency N fertilizers up to 53% when compared to dry granular urea, and up to 35% when compared to liquid urea-ammonium nitrate fertilizers commonly used by farmers. In another study, ARS researchers in Ames, IA tested the use of stabilized nitrogen fertilizer as a means of reducing nitrous oxide (N2O) greenhouse gas emissions from a non-irrigated corn crop; in that case, N2O emissions were not reduced, but crop yield was increased. In the Iowa experiment, the stabilized materials appeared to affect the nitrate and ammonium pools in the soil through a reduction of ammonia volatilization when the materials are applied; the increase in crop yield reflected an increase in nitrogen availability during the grain-filling period as these plants showed a greater amount of leaf greenness, larger grain size and ultimately larger crop yield. Thus, for temperate climates, the soil water balance is a dominant factor affecting N2O emissions, and that changing forms of nitrogen to include stabilized materials could have a yield advantage but not necessarily a greenhouse gas emission advantage. Together, the two studies provide important early steps towards regional-specific guidelines for reduction of greenhouse gases from agricultural sources, while maintaining or increasing crop yields. (NP212 C2 PS-A; PM 6.2.1; #3625-11000-004-00D)

Effects of elevated CO2 on crop yield evaluated. At least nine research groups from around the world have conducted FACE (free-air CO2 enrichment) experiments on at least ten crops over the last two decades to determine how increasing concentration of atmospheric CO2 will likely affect future crop yields. An ARS scientist from the U.S. Arid-Land Agricultural Research Center, Maricopa, AZ analyzed the collective results of these experiments and concluded that yields of sorghum, a C4 crop, were unaffected, whereas yields of C3 crops were increased 16 to 40 percent under atmospheric CO2 concentrations of roughly 550 ppm. For example, yields of C3 grain crops (wheat, rice, barley, soybean) were increased roughly 17 percent on average. Results from the China FACE Rice Project showed 32 percent increases for some hybrid rice varieties. These results show that given minimal climatic change impacts of increasing temperature, changing water availability, etc., future C3, but not C4, crop yield will increase due to the effects of elevated atmospheric CO2 on plants. Given detrimental impacts of increasing temperature, changing water availability, etc., these direct CO2 effects will tend to mitigate the harm. The results are critically important for estimating future food production and offer insights for

potential changes to agricultural systems needed to increase world food production. (NP 212 C3 PS-A; PM 6.2.1; #5347-11000-010-00D)

NP206

Recovering phosphorus from solid manure. ARS scientists in Florence, South Carolina developed a process called "quick wash" to recover phosphorus and reduce its impact in poultry litter. This technology provides an alternative poultry litter management when application onto land is not an option in areas such as Georgia, North Carolina, and Chesapeake Bay. The quick wash technology is comprised of a process to form a concentrated phosphorus solid material, and a washed poultry litter residue containing low phosphorus with most of the original organic carbon and nitrogen. This technology will help the poultry producer to better manage manure and nutrient on their farms. It can facilitate economic phosphorus transport in concentrated form from areas where it is in excess to areas where it is needed for its effective utilization as plant fertilizer. Renewable Organics LLC has applied for exclusive licensing of ARS patent rights. (NP 206, C 2, P.S. 2, P.M 6.2.1, #6657-13630-003-00D)

Benefits of shallow subsurface band application of poultry litter. Poultry litter is typically land-applied by broadcast surface application, but this method has a high potential for undesirable transport of litter nutrients off the field into streams, rivers, lakes, and other bodies of water. An experimental litter application implement that places the litter in shallow subsurface bands in soil was used in five field experiments. When the poultry litter is applied in subsurface bands, compared to traditional broadcast surface application, results show that phosphorus and nitrogen nutrients in water running off field plots are reduced by 80 to 95%. Use of the implement by producers and others who apply litter to fields is expected to reduce pollution of streams, rivers, lakes, and other bodies of water. (NP 206, C 2, P.S. 4, P.M. 6.2.1, #6420-12000-009-00D)

Troughs as a hidden source of Johne's Disease. *Mycobacterium avium* subsp. *paratuberculosis* (*M. paratuberculosis*) causes Johne's disease, a chronic intestinal infection that affects ruminants such as dairy cows. This disease can cause losses of as much as \$200,000 per year in a herd of 1,000 dairy cows. A continued increase in the number of cases of Johne's disease among dairy cattle suggests that there may be uncharacterized sources of contamination on farms. Scientists from the ARS Laboratory in Bowling Green, KY evaluated differences in the ability of the bacteria to adhere to and persist on the surfaces of different livestock watering trough materials. Researchers found high concentrations of *M. paratuberculosis* on all trough materials within three days of inoculating trough water with the bacteria and they persisted for more than 149 days. Persistence of *M. paratuberculosis* was lowest on stainless steel, followed by plastic, galvanized steel and concrete. To reduce exposure of susceptible animals to *M. paratuberculosis* on infested farms, best management practices aimed at reducing slime build-up on trough surfaces should be included in any Johne's control plan. (NP 206, C 3, P.S. 1, P.M. 6.2.1, 6445-12630-003-00D)

Nitrogen use efficiency in dairy production. The more efficient dairy farmers are in managing nitrogen (N), the less N will be wasted in manure. ARS Researchers in Madison, WI, and University Park, PA, along with Australian colleagues, calculated N use efficiency ratings. These ratings could help dairy farmers make better use of their N in the face of escalating costs and increasing nutrient regulation. They found that only about 20-35% of the N fed to dairy cows is converted into milk. They also discovered that 16 -77% of the N in manure or fertilizer is taken up by crops and pasture plants. In addition, the study showed that between 8 and 64% of all N applied to typical commercial dairy farms is converted into farm products. The wide ranges in N use efficiency indicate that there is significant room for improved N management on the average farm, with likely benefits to farm profits and the environment. (NP 206, C 2,

<u>Carbonization of swine solids</u>. High temperature pyrolysis of swine manure and manure blends generates energy co-products similar to low grade coals and natural gas. Pyrolysis is the well known process of heating organic material in the absence of oxygen. It is currently the subject of new study as a method for producing bioenergy and useful byproducts from agricultural biomass. ARS scientists in Florence, SC and an industry cooperator tested high temperature pyrolysis of swine manure and a blend of swine manure and ryegrass. In addition to the pyrolysis oil, the process generated a solid char product with an energy equivalent to that of some coals and a combustible gas with an energy density slightly less than that for natural gas (methane). (NP 206, C 4, P.S. 4, P.M. 6.2.1, #6657-13630-003-00D)

Association of *Enterococcus* species and antibiotic resistance with specific pollutant sources. Enterococci are widely used as indicator of fecal contamination of waterways in most urban areas throughout the United States. Repeated exposure of these bacteria to antibiotics may result in antibiotic resistance and become a serious public health threat. This research was conducted at the USDA ARS Salinity laboratory in Riverside, CA to investigate the influence of the apparent source of *Enterococcus* in waterways on their resistance to a number of antibiotics. *Enterococcus* species showed multiple resistances to ciprofloxacin, erythromycin, and tetracycline. Resistance to tetracycline was most prevalent in samples collected from sediments that were impacted by agricultural activities, while ciprofloxacin and erythromycin resistance were prevalent in samples impacted by urban runoff. This information will provide useful information for water utilities on microbiological pollutant types and sources affecting the Santa Ana River and can help the development of best management practices for water quality improvements. (NP 206, C 3, P.S. 2, P.M. 6.2.1, #5310-32000-002-00D)

Drinking water treatment residuals reduce selenium. Selenium is a naturally occurring trace element commonly found in soils in the western U.S. selenium can cause reproduction failure, deformities, and die-off of migratory waterfowl, fish, insects, and plants. Drinking water treatment residuals are a solid byproduct, composed primarily of alum that is used to remove very small solid particles from the water supply. Collaborative research involving scientists from ARS in Kimberly, ID, Colorado State University, EPA National Risk Management Research Laboratory, and Argonne National Laboratory tested the ability of drinking water treatment residuals to reduce selenium in water. In this laboratory study, drinking water treatment residuals were added to selenium solutions. Results indicate that water treatment residuals can reduce waterborne selenium concentrations, and the adsorbed selenium is stable and not likely to be released back into the environment. Drinking water treatment residuals, which are a normal byproduct of drinking water treatment, can potentially be used to remove soluble selenium from areas impacted by selenium such as Kesterson Reservoir and the Salton Sea in California. (NP 206, C 4, P.S. 3, P.M. 6.2.1, #5368-12630-002-00D)

Manure nutrient losses are reduced by stiff-stemmed grass hedges. ARS scientists in Lincoln, Nebraska measured the effectiveness of narrow grass hedges in reducing runoff nutrient transport from sites on which beef cattle manure was applied to meet 0, 1, 2 or 4-year corn phosphorus requirements. Manure application rate significantly affected the transport of phosphorus in runoff on the treatments without a grass hedge. However, phosphorus transport on the treatments where manure was applied to plots containing a grass hedge was similar to the treatments that did not receive manure. This research indicates that stiff-stemmed grass hedges reduce the transport of nutrients in runoff occurring soon after manure application, and an adoption of grass hedges would decrease phosphorous runoff into streams and enhance water quality. (NP 206, C 2, P.S. 4, P.M. 6.2.1)

Non-composted municipal solid waste (MSW) processing byproduct improves U.S. Army training land reclamation. The costs to the U.S. Army for handling, management, and disposal of non-hazardous solid waste are significant and are expected to increase. A new MSW processing technology has been

developed that sterilizes and separates the MSW and produces a light cellulose pulp called Fluff®. The U.S. Army has large areas that have been damaged due to extensive training activities. These areas often lack sufficient topsoil, organic matter, and nutrients necessary for successful revegetation. A study was conducted at Fort Campbell, KY, to evaluate the Fluff as a soil amendment for improving soil quality, plant growth, and revegetation success on training lands. The highest Fluff rate improved native grass establishment. Plant phosphorus (P) accumulation also increased significantly with increasing Fluff application. Because no adverse environmental effects were detected and Fluff improved perennial grass establishment, MSW processing followed by land application of the resulting Fluff should be considered to be a viable and beneficial alternative to current waste management and revegetation practices. (NP 206, C 4, P.S. 3, P.M. 6.2.1)

Nitrogen and phosphorus losses from beef cattle feedyards. Losses of nitrogen and phosphorus from animal feeding operations can potentially have adverse effects on the environment; however, the utilization and losses of these nutrients from beef cattle feedyards need to be better understood. Scientists at the ARS Conservation and Production Research Laboratory, Bushland, Texas, conducted a nitrogen and phosphorus balance study at two large (greater than 20,000 head capacity) beef cattle feedyards in the Southern Great Plains. On average, approximately 31% of fed nitrogen was excreted in feces and 55% was excreted in urine. Over time, 48% of the excreted nitrogen was retained in the manure pack and 52% was apparently lost to the atmosphere via volatilization. However the values were greatly affected by diet and season. These data will be useful for nutrient management planners in preparing nutrient management plans for beef cattle feedyards and also in evaluating nutrient losses from feedyards. (NP 206, C 2, P.S. 1, P.M. 6.2.1)

NP215

Expression of a red clover gene in alfalfa could result in major savings of nitrogen. ARS researchers at Madison, WI previously identified a novel enzyme in red clover that produces phaselic acid. Phaselic acid accumulates to high levels in red clover and is a key factor in a natural system of post-harvest protein protection in red clover that reduces the loss of nitrogen. Phaselic acid also appears to provide ultraviolet (UV) protection and other plant defenses. The researchers have transferred the gene encoding this enzyme to alfalfa, which does not normally accumulate phaselic acid. The resulting plants made small amounts of phaselic acid, along with larger amounts of the related compounds, p-coumaroyl- and feruloyl-malate. This finding strongly suggests the potential for increasing levels of phaselic acid in other forage crops like alfalfa to improve post-harvest protein protection. If alfalfa could achieve the level of protection found in red clover, farmers would save more than \$100 million annually in lost nitrogen while substantially reducing levels of environmental nitrogen waste from ruminant animal production systems. (NP215 C3 PS-H; P.M. 6.3.1, #3655-21000-046-00D)

Measures of ecosystem health and trends. Science-based measurements of the impacts of management practices on rangeland ecosystems are needed to select cost-effective conservation practices for the nation's hundreds of millions of acres of public and private rangelands. Long-term, accurate, affordable measurements of selected characteristics of rangeland soils, plants and animal life provide indicators of status and trends in ecological health. ARS scientists at Las Cruces, NM worked closely with NRCS and other scientists to develop new, improved vegetative measurements and soils field tests to aid in assessing rangeland health. These measurements were incorporated with data from the National Resource Inventory to assess resource status across the western US. A key finding was that non-native species currently occur on nearly 50% of non-federal rangelands, and account for at least 50% of plant cover on over 5% of these private lands. These soil and vegetative measures are now being used to significantly improve the assessment of the susceptibility of rangeland soils to wind erosion. These improved tools will help

private landowners along with public land managers assess conservation needs and select appropriate practices. (NP215 C1 PS-A; P.M. 6.3.1, #6235-11210-006-00D)

Yellow bluestem pastures don't benefit from prescribed burn or herbicide treatments. Yellow bluestem pastures are important for livestock production in the southern Great Plains. They are often burned or sprayed with herbicides annually to control weedy species in the belief that these treatments will increase forage production and livestock performance. However, producers were concerned with whether the expense and environmental risk of these costly practices is justified the ARS scientists at Woodward, OK, in collaboration with Kansas State University, evaluated these practices and found that regular prescribed fire or herbicide applications in the spring are not necessary for the optimal management of Yellow bluestem pastures and do not increase livestock performance. Adequate fertilization and proper grazing management are more effective annual treatments. Eliminating these ineffective practices will result in a substantial savings while reducing the risks associated with burns and herbicide usage. (NP215 C1 PS-B; NP215 C2 PS-F; P.M. 6.3.1, #6216-21630-008-00D)

Comparing cumulative-germination responses of cheatgrass and five native perennial grasses. Cheatgrass now dominates millions of hectares of rangeland in the western United States by outcompeting native vegetation following fire. ARS scientists at Boise, Idaho studied the germination response of cheatgrass and 5 different native bunchgrasses by developing a model simulating potential germination response for any set of weather conditions occurring over 38 years. By comparing potential response over such a long time period, the researchers confirmed that on average, cheatgrass is 2-5 times faster than other species in early germination. Their modeling techniques also identified the types of weather conditions necessary to successfully establish the more desirable native species plant as part of an integrated strategy to control cheatgrass. With this improved understanding of climatic requirements for establishment of different species, land managers can make more cost-effective decisions about re-vegetation and restoration practices in a given year. (NP215 C1 PS-A; P.M. 6.3.1; #5362-13610-009-00D)

Native grasses from Northeastern CRP lands for biofuel production. Concerns about finding sufficient land for biofuel production have led to the evaluation of Conservation Reserve Program (CRP) lands as potential production sites. An extensive study by ARS scientists at University Park, Pennsylvania of grassland sites across major northeastern ecoregions determined the effects that plant species composition, diversity, above ground biomass, and chemical composition had on potential biofuel yield. The study found that CRP lands with a high proportion of native warm-season prairie grasses have the potential to produce more than 600 gallons of ethanol per acre while still maintaining the ecological benefits of grasslands. (NP215 C2 PS-D; P.M. 6.3.1; #1902-21000-007-00D)

Forage kochia increases livestock carrying capacity of western rangelands. Low forage production and poor nutritional content of western rangelands limits their value for fall and winter grazing. ARS scientists in Logan, Utah, in cooperation with animal scientists at Utah State University, compared the carrying capacity and livestock performance of traditional winter pastures dominated by crested wheatgrass and cheatgrass (an invasive annual weed) versus similar rangelands seeded with crested wheatgrass and forage kochia. The rangelands with forage kochia had higher crude protein, and increased carrying capacity by six-fold when compared to the non-treated rangelands. Cattle on rangelands with forage kochia had higher increases in body condition than cattle on the crested wheatgrass/cheatgrass rangelands. This research demonstrates that seeding western rangelands with forage kochia can improve sustainability of livestock production in the western U.S. by providing increased amounts of nutritious forage during the fall and winter. (NP215 C2 PS-G; P.M. 6.3.1; #5428-21000-012-00D)

<u>Shrubs in northern grasslands increase carbon and nitrogen in soil.</u> Overgrazing and fire suppression have contributed to an increased abundance of shrubs within grasslands throughout the world. Increases in shrubs have come at the expense of grasses, but little is known about how this shift in vegetation

dominance will affect grassland ecosystem functions. ARS scientists in Mandan, ND conducted a study to determine the influence of shrub expansion on soil carbon and nitrogen in northern mixed-grass prairie grassland. They found that carbon and nitrogen were greater under established shrubs than under grassland in the surface six inches of soil. Accumulation of soil carbon and nitrogen under shrubs in northern grasslands across the world may contribute to 'islands of fertility' that reduce greenhouse gases while offering opportunities to manage these grasslands for more sustainable livestock production. (NP215 C2 PS-F; P.M. 6.3.1; #5445-21310-001-00D)

<u>Using growth regulator herbicides to control rangeland weeds.</u> Growth regulator herbicides are commonly used to control broadleaf weeds on croplands and rangelands. For cereal crops, however, these herbicides often reduce cereal yield if applied to the plants while they are developing seeds. ARS scientists at Miles City, MT, tested whether 2,4-D, dicamba, and picloram at typical field use rates would reduce seed production of Japanese brome, an invasive weed. They found that that picloram reduced seed production nearly 100% when applied at the internode elongation, boot, or heading stages of growth, whereas dicamba appeared to be slightly less effective and 2,4-D was much less effective. Results were corroborated in the field where typical use rates of aminopyralid and picloram reduced Japanese brome seed production by more than 95% when applied at three different plant growth stages. These results indicate that using growth regulator herbicides to reduce seed production, and thereby depleting its short-lived seed bank, can be part of an integrated strategy to help control Japanese brome. NP215 C1 PS-C; PM 6.3.1, #5434-21630-002-00D)

Water savings from conservation tillage of peanut and cotton. ARS scientists at Dawson, GA showed that the use of conservation tillage for peanut and cotton lowered crop water usage by 20% versus conventional tillage while crop yield and quality were maintained. Conservation tillage also resulted in lower emissions and lower production costs. (NP 216, C3, PS 3A; PM 2.2.1; #6604-64000-007-00D) Cover crops to maintain soil carbon. Maintaining soil carbon and soil productivity is especially important in cropping systems from which crop residues are harvested for corn silage production or biofuel production. ARS researchers in Ames, IA found that winter rye cover crops maintained soil carbon content in a corn silage-soybean rotation, whereas soil carbon decreased by 6% without the cover crops. Use of cover crops can permit the harvesting of corn stover for animal feed or biomass without depleting soil carbon or soil productivity. (NP 216, Component 1, Problem Statement 1A; PM 2.2.1 #3625-21610-001-00D)

NP216

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Cover crops to maintain soil carbon. Maintaining soil carbon and soil productivity is especially important in cropping systems from which crop residues are harvested for corn silage production or biofuel production. ARS researchers in Ames, IA found that winter rye cover crops maintained soil carbon content in a corn silage-soybean rotation, whereas soil carbon decreased by 6% without the cover crops. Use of cover crops can permit the harvesting of corn stover for animal feed or biomass without depleting soil carbon or soil productivity. (NP 216, Component 1, Problem Statement 1A; PM 2.2.1 #3625-21610-001-00D)

Annual forages are profitable alternatives to summer fallow in semi-arid dryland farming operations. Economics are driving the replacement of wheat-summer fallow cropping systems by diversified and continuous (every year) cropping systems in dryland crop production areas of the semi-arid Northern Great Plains. A five year study conducted by ARS scientists at Sidney, Montana showed that replacement of the summer fallow with annual spring-seeded forage crops resulted in less water and nitrogen for the following wheat crop. Durum grain yields were reduced by 10.8 bushels per acre. However, annual forage yields averaged 2.5 tons per acre and greatly reduced herbicide use resulting in annualized net returns in the three annual forage-durum systems of \$51 (forage barley), \$31 (forage barley inter-seeded with field pea), and \$14 (foxtail millet) per acre greater than for fallow-durum. (NP216 C1 PS-A; NP216 C4 PS-A; NP211 C1 PS-A; PM 2.2.1; #5436-13210-005-00D)

Minimal tillage practices substantially reduce sugarbeet production costs. Sugarbeet growers in the Northern Great Plains need to reduce the cost of energy, labor and time invested in farming while increasing profits. Tillage practices are a major expense. A 4-yr field study conducted by ARS scientists at Sidney, MT showed that farmers could substantially reduce tillage costs using minimal tillage practices (i.e., strip tillage) with little significant effect on sugarbeet yield or quality. Strip tillage had no significant effect on root yield, sucrose content, or sucrose yield, and, unlike no tillage, seedling emergence was not significantly impacted. As a result of this and related studies, sugar beet producers in the Northern Great Plains and in other regions have begun incorporating strip tillage in their operations in ever increasing numbers because of the economic benefits demonstrated over no-till and deep tillage, which requires heavier equipment and higher costs. (NP 216 C1 PS-A; NP 211 C1 PS-B; NP211 C1 PS-E; PM 2.2.1; #5436-13210-005-00D)

Sheep grazing can control weeds and increase nutrient cycling in soils to sustain crop yields. Wheat and livestock producers are looking for ways to reduce herbicide and feed costs, respectively. The largest use of glyphosate herbicides in Montana is for control of volunteer wheat during fallow periods. In a five-year study conducted in collaboration with Montana State University, ARS scientists at Sidney, MT, found that grazing had no effect on wheat grain and biomass yields, but reduced the need for herbicides by reducing populations of some problem weed species. By reducing the amount of crop residue returned to the soil compared with tilled and no-tilled treatments, grazing slightly reduced soil carbon, pH and available nitrogen, phosphorus; however soil calcium, magnesium, and sodium contents increased. The study demonstrated that properly timed sheep grazing can be an effective way to economically control volunteer wheat and other weeds and sustain crop yields by reducing selection pressure for weed resistance to glyphosate, decreasing potential leaching of nitrates, and increasing nutrient cycling in soils. (NP216 C1 PS-A; PM 2.2.1; #5436-13210-005-00D)

Long-term use of poultry litter and conservation tillage: Agronomic and environmental assessment. Conservation tillage and animal manures as fertilizers can make significant contributions for sustainable food production but their interactions need to be studied over long periods of time to test for possible negative impacts. Scientists from the USDA-ARS, in Watkinsville, GA, and the University of Georgia, Athens, Georgia examined 14 years of research in crop productivity, soil quality, and water quality associated with the use of conservation tillage and poultry litter in cotton and corn production. Yields of cotton and corn increased on average 10-27% with no-till and 32-42% with combination of no-till and poultry litter, but soil nutrient accumulation, particularly phosphorus and zinc, reached excessive levels from long-term use of poultry litter. These results can be used by State Cooperative Extension Systems, USDA-NRCS, and other state and federal agencies to guide producers in management of systems with conservation tillage and an organic source of fertility such as poultry litter. (NP 216, C1, PS 1; PM 2.2.1; #6612-61660-002-00D)

Feather meal and feather meal-poultry litter blends show potential as side-dress fertilizers to improve nitrogen use efficiency in organic systems. Improving nitrogen use efficiency in corn production will

provide economic benefits to farmers and environmental benefits to society. Researchers at ARS in Beltsville, MD showed that the N release characteristics of locally available sources of Organic Materials Review Institute (OMRI)-approved N (raw poultry litter, pelletized poultry litter, feather meal, and a pelletized blend of feather meal and poultry litter) could serve as viable sources of supplemental N for organic and other grain production. The rate of N release of each material was relatively fast, suggesting that applying them to corn at the V5 growth stage (sidedress) could result in significant improvements in N use efficiency compared to pre-plant applications (the industry standard). Results showed that the feather meal and the feather meal-poultry litter blend were more nutrient dense than the raw and pelleted poultry litter and therefore less costly to transport per unit of available N. This information will benefit producers, extension educators, nutrient management plan writers, and others interested in improved soil fertility and nutrient management in organic and other grain cropping systems. (NP 216, C1, PS 2; PM 2.2.1 #1265-21660-003-00D)

Potato simulation model incorporates effects of increased atmospheric carbon dioxide. Mathematical crop models have been developed over the past forty years for farm management and crop inventory tools. Despite significant improvements, the models still need to incorporate the effects of increased atmospheric carbon dioxide associated with climate change on plant growth. Scientists in Beltsville, MD developed and tested an improved mathematical model of photosynthesis for potato that is responsive to sunlight, temperature, and atmospheric carbon dioxide. The improved photosynthesis model was merged with other recent research results into a new potato growth simulation model (SPUDSIM) and tested over a wide range of conditions with good results. The increased accuracy of the model will enable producers to better manage production, and will enable scientists and policy-makers to make better assessments of food production under conditions of changing climate. (NP 216 C4 PS-D; PM 2.2.1; #1265-61660-006-00D)

Crop rollers terminate cover crops and reduce pesticide-use. An integral component of conservation agriculture systems in cotton is the use of a high-residue winter cover crop. However, planting into high-residue is challenging and terminating these cover crops with herbicides is costly. ARS scientists at Auburn, AL developed a reduced-herbicide method for terminating cover crops. Black oat, rye, and wheat winter cover crops were flattened with a straight-blade mechanical roller-crimper alone, or followed by four reduced rates of herbicide application. Results showed that rolling followed by herbicide applied at rates less than half the standard application rate effectively and reliably terminates mature cereal winter cover crops, thus maintaining cotton population and protecting growth. Rolling mature winter cereal cover crops will also likely conserve more soil moisture compared to standing cover. However, rolling immature cereal cover crops provides no benefit. When put into widespread practice, the roller-reduced herbicide application will decrease agrochemical use, thus leading to reduced farming impacts on the environment, and increased producer profit margins. (NP 216 C1 PS-A; PM 2.2.1; #6420-12610-

NP308

Effective methyl bromide alternatives demonstrated in production of raspberry nursery stock. Raspberry nurseries have relied on pre-plant fumigation with methyl bromide to produce approximately seven million plants annually for an annual fruit crop worth about \$278 million. Methyl bromide is used to control plant pathogens, weeds, and nematodes which would severely reduce the quantity and quality of the crop, but use of this fumigant is being phased out worldwide under the Montreal Protocol. ARS researchers in Davis, California tested fumigant alternatives to methyl bromide, including Inline® (1,3-D+chloropicrin formulated for drip application), Telone C35® (1,3-D + chloropicrin, shank-applied), Midas® (iodomethane + chloropicrin, shank-applied), and plastic mulch sealing films at an experiment station and three commercial nurseries in California and Washington. These combinations emerged as effective alternatives to methyl bromide and chloropirin, and matched the methyl bromide standard for

plant production and pest and pathogen control. Virtually impermeable film (VIF) generally improved efficacy of the alternative fumigants compared to standard high density polyethylene, but solarization was not effective. Transition from methyl bromide to Midas®, although technically feasible, may be complicated by uncertain registration status and relatively high material cost. However, Telone C35®, especially in combination with VIF, was validated as an effective methyl bromide alternative for raspberry farmers and nurseries. (NP308; C1, PS 1A, 1B, 1C, & 1D; PM 4.2.3; Project # 0500-00044-021-00D)

Low permeable films reduce fumigant emissions. In comparison with the use of standard polyethylene films, the use of low permeability films enables lower doses of fumigant to be used and also reduces the amount of fumigant, which might cause unintended environmental damage. A new low permeable film, referred to as totally impermeable film or "TIF" effectively reduced fumigant emissions in laboratory tests. ARS scientists in Parlier, California conducted a large field trial to test the new film for reducing fumigant emissions. The TIF peak emission rate was as much as 10 times lower than that from standard polyethylene (PE) film. Over a six day field covering period, the total emission loss with TIF was reduced below two percent of total fumigant applied, compared to 30 percent emission with the PE film. However, the emission surge upon cutting the TIF tarp was much higher than with the PE film, indicating that a longer waiting time would be needed with TIF to reduce potential exposure risks. The research showed that using this new film will help improve buffer zone restrictions and enable many fields to be fumigated under the newly amended EPA regulations. (NP308; C1, PS 1B, 1C, & 1D; PM 4.2.3; Project # 5302-13220-004-00D)

New Web-based database on *Phytophthora* genus will aid in worldwide disease management. The fungal genus *Phytophthora* is responsible for many diseases of crop plants worldwide and consists of approximately 106 species which are difficult to identify. ARS scientists with university collaborators have developed a Web site to support research on *Phytophthora* that includes complete morphological descriptions, information on host range and geographical distribution, a comprehensive molecular phylogeny using seven nuclear genes, and a section on molecular identification and detection. Expansion of the database Web portal to include the related Oomycete pathogens Pythium, downy mildews, and Albugo was also initiated. This database will serve as a resource for researchers working on the genus, as well as a repository for future work. The Web-based database will enhance the understanding of the *Phytophthora* genus, simplify species identification, and stimulate further research on the genus in aid of disease management. (NP308; C1, PS 1D; PM 4.2.3; Project # 5305-22000-012-00D

Alternatives to methyl bromide demonstrated in key crop systems and regions formerly dependent solely upon methyl bromide. Key crop systems have relied solely on fumigation with methyl bromide for pest and pathogen control, but use of this fumigant is being phased out worldwide under the Montreal Protocol. ARS scientists in Fort Pierce, Florida, along with university collaborators, conducted 48 large-scale field demonstration trials, using the best available, industry-appropriate alternatives to methyl bromide. The alternatives included substitute fumigants and supporting integrated pest management (IPM) practices. Trials were conducted in partnership with commercial growers at sites adequately representing the biological and environmental diversity of the production systems of tomato, pepper, eggplant, strawberry, forest nursery seedlings (loblolly pine), sod, ornamentals (caladium), and cut flowers (delphinium). Grower demonstration trials were performed on all of those commodities and turf and cucumber in Alabama, Florida, Georgia, South Carolina, and North Carolina. The size of each grower demonstration trial ranged from one-half acre to 58 acres. These trials demonstrated that technically feasible alternatives to methyl bromide soil fumigation are available and can control pests adequately without the environmentally damaging effects of methyl bromide. (NP308; C1, PS 1D; PM 4.2.3; Project # 0500-00044-020-00D)

Novel red flour beetle trap developed. The red flour beetle is a major pest of stored food products such as flour. Effective pest monitoring is critical to pest management systems, such as those proposed as alternatives to methyl bromide, because they provide necessary guidance for timing and targeting to the application of control measures. ARS researchers in Gainesville, Florida, have developed a new trap for monitoring the red flour beetle that uses ultraviolet light, a chemical attractant, and a physical configuration that guides beetles into a pitfall. In small scale experiments, trap efficiency was estimated to be about 33 percent, i.e., the trap captured one third of the flour beetles present. This is a comparatively high efficiency with promise as a monitoring tool for use in flour mills and other food processing plants. (NP308; C2, PS 2A; PM 4.2.3; Project # 6615-22430-003-00D)

New information for the management of nematodes and other soilborne pests in floriculture production systems discovered. Root-knot nematodes are major pest for cut flower production in the southeastern United States. ARS researchers in Fort Pierce, Florida, in collaboration with faculty at the University of Florida, developed new information on the susceptibility of a variety of important cut flower species to two common species of root-knot nematodes in Florida. Experiments evaluating the susceptibility of selected flower cultivars to the root-knot nematode species *Meloidogyne incognita* (race two) and *Meloidogyne Javanica* (race one) revealed that snapdragon and nasturtium were susceptible to and supported high populations of both species, while marigold, zinnia, salvia, and carnation cultivars were not susceptible nor good hosts. Knowledge of crop nematode susceptibility will be used for managing nematode populations with crop rotations. (NP308, C 1, P.S. 1C, P.M. 4.2.3, Project #6618-22000-036-00D)

Oxygen increases toxicity of phosphine to insect pests. Phosphine is a slow acting fumigant in controlling insects, and treatment time can last over 10 days for some insects, limiting the usefulness of this fumigant as a quarantine treatment. For low temperature phosphine fumigation of fresh commodities, treatment is even longer as the toxicity of phosphine decreases at lower temperatures. Shortening treatment would increase turnover time of fumigation chambers and reduce fumigation cost. ARS researchers in Salinas, California, found significant synergistic effects of oxygen in increasing the toxicity of phosphine against all life stages of various insect pests which resulted in significant reductions in treatment time for pest control. The synergism may have significant practical implications in developing more effective and shorter fumigation treatment for postharvest pests especially for low temperature phosphine fumigation on perishable commodities. (NP308; C2, PS 2A; PM 4.2.3; Project # 5305-43000-003-00D)

Quarantine strategies to control Hessian fly in exported hay developed. United States and foreign regulatory agencies are seeking new methods to ensure that the Hessian fly is not accidentally introduced to new areas through hay shipped from the western states. ARS researchers in Parlier, California, examined different methodologies to control this insect pest. Hay harvesting and drying practices increased mortality of Hessian fly puparia in warm and arid climates where export quality hay is grown. In addition, fumigation with a phosphine and carbon dioxide gas mixture completely controlled this pest in laboratory tests. This work supports the concept that the occurrence of Hessian fly in harvested, processed, and fumigated hay bales is negligible and protects a \$660 million annual foreign market. (NP308; C2, PS 2B; PM 4.2.3; Project # 5302-43000-033-00D)

Management Initiative 2: Ensure Provision and Permanent Access of Quality Agricultural Information for USDA, the Nation, and the Global Agricultural Community via the National Agricultural Library

Select Examples of Recent Progress:

The National Agricultural Library (NAL) is the largest and most accessible agricultural research library in the world. It provides service directly to the staff of USDA and to the public, primarily via the NAL Web site, http://www.nal.usda.gov. NAL was created with the U.S. Department of Agriculture (USDA) in 1862 and was named in 1962 a national library by Congress (7USC§3125a), as "the primary agricultural information resource of the United States." NAL is the premier library for collecting, managing, and disseminating agricultural knowledge. The Library is the repository of our Nation's agricultural heritage, the provider of world-class information, and the wellspring for generating new fundamental knowledge and advancing scientific discovery. It is a priceless national resource that, through its services, programs, information products, and Web-based tools and technologies, serves anyone who needs agricultural information. The Library's vision is "advancing access to global information for agriculture."

Progress towards becoming "Digital NAL". For decades, NAL has delivered some services and content digitally. The goal of "Digital NAL" is to deliver information about all NAL programs and services digitally and to deliver as much content and as many services digitally as are permitted by law, technology, and funding. Because NAL's resources are limited, its customer needs and expectations are broad, and the digital information landscape is constantly changing, practical steps have been identified to advance the development of "Digital NAL." A number of projects to re-engineer NAL operations and improve NAL's digital services will be completed in FY2011. In addition to projects to increase and improve retrieval of information from NAL's Web site and databases, the Library's information technology infrastructure is being reinforced and brought up to date. The NAL collection development policy is being revised. The DigiTop and DigiCALS services for USDA personnel are being combined and re-engineered. Growth of the AGRICOLA index has been identified as a critical immediate task required to further a Digital NAL This will involve significantly increasing the number of articles indexed annually from ~75,000 to 180,000. To accomplish this, a fundamental transformation is necessary from human generated index entries to automated indexing using specialized computer applications. The process has started for acquiring an automated indexing application, expanding the NAL thesaurus, and adjusting the indexing workflow. The transformation will be completed in FY 2011. The indexers will become editors performing quality control and systems tuning with a goal to increase annual production to \sim 300,000 articles in the next few years.

<u>Delivering information and research services</u>. NAL provides free access to agricultural information, primarily through its core Web site, <u>www.nal.usda.gov</u>. NAL's FY2010 total volume of direct customer transactions exceeded 90 million transactions. Services delivered digitally continued to grow while services based entirely on physical materials continued to decline. Examples of accomplishments and progress include:

- DigiTop usage increased while document delivery services decreased. USDA staff executed 1,353,306 full text downloads from NAL's DigiTop (Digital Desktop for USDA) service, posting a 7.5% increase in downloads from FY2009. In contrast document delivery requests received by NAL decreased by 20 percent (34,469) as more material became available online. 100% of document delivery requests were received electronically and 84% delivered electronically. Reference transactions increased 15.6% to 15,451.
- Outreach and instruction increased. There was a 13.8% increase in presentations and a more than 100% increase in the number of customers reached. NAL staff exhibited at numerous events and

conferences to increase visibility of NAL services and products. Venues included: Future Farmers of America annual conference; USDA People's Garden; USDA Earth Day; USDA Agricultural Festival; USDA National Nutrition Month; Preservation Week; Charles Valentine Memorial Lecture; Sustainable Agriculture & Food Systems Assoc.; Joint Annual Meetings of the American Dairy Science Assn., Poultry Science Assn., American Society of Animal Science, Asociacion Mexicana de Produccion Animal, and the Canadian Society of Animal Science; 20th Annual Social Marketing in Public Health Conference; and the Food Safety Education Conference. NAL continues to look strategically at how it can increase its reach by targeting high impact events.

- NAL's AFSIC received project funds to support organic livestock producers and researchers. The
 Alternative Farming Systems Information Center received \$114,000 from USDA's National
 Institute of Food and Agriculture and Agricultural Marketing Service to select, digitize, and make
 publicly available important USDA publications on organic livestock production.
- NAL received project funds to support digital conversion of the USDA Pomological Watercolor Collection. The Library received \$206,600 from The Ceres Trust to fund the conversion of more than 7,000 original watercolor paintings of the USDA Pomological Watercolor Collection.
- NAL's AFSIC/RIC received \$1.5 million grant to help beginning farmers and ranchers start and stay in farming and ranching. NAL's Rural and Alternative Farming Systems Information Center Information Centers, in partnership with the American Farm Bureau Federation, were awarded a five-year grant to create and implement a National Curriculum and Training Clearinghouse, called http://www.Start2Farm.gov.
- NAL receives renewed funding to digitize Fruit and Vegetable Market News Reports collection. Funding received from USDA/AMS since 2006 has supported digitization of over 2,200 titles (~78,800 pages) of AMS Reports which provide detailed marketing information for fruit and vegetable commodities at domestic wholesale markets and production areas, since 1916.
- Michelle Obama's www.LetsMove.gov . NAL's Food and Nutrition Information
 Center/Nutrition.gov specialists provided advice and digital content to populate the Let's Move:
 America's Move to Raise a Healthier Generation of Kids Web site, launched in February, from
 NAL's SNAP-ED Connection's Recipe Finder database, Food Assistance Resource List, and
 Healthy Meals Resource System. The content supplied by NAL supports the First Lady's four
 pillars aimed at solving the childhood obesity epidemic within a generation.
- Directory of Chefs Partnering with Let's Move! The NAL Food and Nutrition Information Center's Directory of Chefs was selected by the White House to use as the basis for the First Lady's Chefs Move to Schools Web site. The Directory matches chefs with schools to improve meals, enhance food workers' skills and teach children about better nutrition. Working under a very short timeline, NAL staff made improvements and enhancements to the existing database. Two new features were created: a geographic display; and a browsable list by state. The May launch was followed by other events which featured the new directory and encouraged new registration. As of the end of FY 2010, over 1897 chefs and more than 1415 schools had signed up to participate.
- NAL supports USDA's Know Your Farmer Know Your Food Initiative. NAL provides staff to
 triage and respond to reference requests which come to the KYF2 site and helped create a
 seasonal poster series promoting Farm-to-Table healthy eating and eating local. The posters and
 associated educational materials complement the KYF2 concept and promote access to NAL
 food, nutrition, and sustainable agriculture information services.
- NAL's Food and Nutrition Center developed a new online tool that calculates an individual's daily nutrient recommendations based on the Dietary Reference Intakes (DRI). The Interactive DRI for Healthcare Professionals, http://fnic.nal.usda.gov/interactiveDRI, available for registered dietitians, doctors, nurse practitioners, and others interested in dietary planning, resulted from collaboration with the DHHS Office of Disease Prevention and Health Promotion and was featured at the American Dietetic Association's Food and Nutrition Conference and Expo.

- Safety Net Preservation Workshop. A workshop entitled "Can We Relax Yet? Assessing the Risks to Library Collections and Operations" was hosted by NAL in June. Library professionals learned practical approaches to risk assessment and risk management models for collections. The NAL building was used for the workshop exercise; as a result, valuable feedback was received for action. The workshop was provided in support of a cooperative disaster response network of Federal libraries and was co-sponsored by NAL, the Library of Congress and LYRASIS.
- When Beans Were Bullets: War-Era Food Posters. On June 21, NAL opened a new exhibit of food-themed posters from the World War I and World War II eras. Cory Bernat, now a project archivist at the National Park Service, independently developed the exhibit based on research she did for her Master's thesis at the University of Maryland, uncovering the educational and patriotic gems among unprocessed posters within NAL's Special Collections. Venues for the exhibit include: NAL; Whitten Building; South Building Cafeteria; and FDA. An online version is available at: http://www.good-potato.com/beans_are_bullets/index.html. Reproductions of popular posters were developed for sale by NAL.
- Heirloom Apple Event Focuses on Diversity and NAL Collections. On September 17, PSD hosted a roundtable discussion on heirloom apples that drew over 100 people from across the Washington area. Seven noted apple experts addressed the very serious issue of dwindling apple varieties and the steps being taken to preserve them. NAL's unparalleled collections in pomology helped shape and inform their research over the years. Speakers from ARS and NIFA provided context, addressing the important relationship between USDA and private growers and the role of USDA in apple research. PSD staff mounted an exhibition of heirloom apples varieties from the USDA Pomological Watercolor Collection, produced a special bibliography, and reproduced images for sale.
- NAL's Food Safety Information Center partnered with United Kingdom's Microbiological Safety of Food Funders Group (MSFFG). The partnership will enable new international food safety research project records to be added to the Research Projects Database at the NAL Food Safety Research Information Office (FSRIO). The initial load included 459 new projects.
- Integration of DigiTop and Current Awareness Literature Service. Mark Logic software system was procured to integrate the Current Awareness Literature Service and DigiTop services beginning January 2011. Cost savings and a more robust search and discovery platform for USDA staff are among the expected outcomes.
- NAL tests reference transaction management software. The Research and Reader Services Team completed pilot testing of RefTracker a commercial software product that integrates processing, response and statistical data management for reference transactions by library research staff. A Decision on acquisition will be made in FY 2011.
- Reorganization of NAL's Public Services Division. NAL completed a major divisional restructuring that is scalable and reflects current workflows, synergies, fiscal realities and opportunities for growth. A major benefit is better alignment of functions to support customer services.

Developing decision support tools for science-based sustainability practices. Concern for the environment has increased consumers' interest in how agricultural products are grown and made. As a result, researchers and the food industry are working to better assess the environmental impacts of processes and activities that span an agricultural product's life cycle, from the acquisition of raw materials to the product's eventual disposal. Currently, however, few information resources devoted to this life cycle inventory exist, particularly for products originating in North America. NAL has begun an initiative to build a database of data sets gauging the material and energy inputs of production processes, along with the outputs released to the environment during production, use and disposal. This life cycle assessment database can then be expanded to include data from other industries, so that one can evaluate the potential environmental impacts of products throughout their life cycle and the processes in place to produce them.

Such data can then inform decisions about changing processes or the materials that go into them to reduce the burden on the environment. NAL expects a test set of data to be ready by spring of 2011.

<u>Building a digital repository.</u> During FY 2010, NAL added approximately 10,000 items to its digital repository. By the end of FY 2010, NAL's digital repository included about 42,000 items; about 750 items are added each month. In June, 2010, the Library began a re-engineering project to unify the existing three platforms into a single platform with a simplified search interface and a streamlined process for adding items and descriptions. This project should be completed in June 2011.

Enriching AGRICOLA. At the end of FY 2010, AGRICOLA included 4,937,064 records of which 1,004,968 were online catalog records and 3,932,096 were indexing records. NAL added 5,814 cataloging records and 34,312 indexing records in FY2010. This is a slower pace than previous years and reflects the reallocation of indexers from production to implementing automated indexing as well as the end of funding for contract cataloging projects.

Developing public awareness and partnerships.

- AgNIC: NAL serves as the secretariat for the Agriculture Network Information Center (AgNIC) Alliance, a voluntary, collaborative partnership that hosts an international distributed network of discipline-specific agricultural information Web sites (http://www.agnic.org). AgNIC provides access to high-quality agricultural information selected by its 62 AgNIC partners, including landgrant universities, NAL, and other institutions around the world. The AgNIC Alliance continues to improve the information technology that supports the AgNIC portal. During 2010, the focus was to build content. The AgNIC search now incorporates 2/3 of the AGRICOLA database and 1.5 million PubMed records which include links to local libraries owning the items, in case people would like to borrow them locally. Along with Web 2.0 services, AgNIC harvests over 30 relevant full-text digital repositories from institutions worldwide, in multiple languages, with the number of repositories harvested ever increasing. During the first six months of the year, AgNIC launched an Animal Health Portal, http://animalhealth.agnic.org/, working with Oklahoma State University and Washington State University. This portal utilizes the AgNIC technologies which are all Open Source, or freely available. AgNIC accepted two new partners during the year: Fort Valley State University, to create a comprehensive collection of information on "goat meat", and SANREM CRSP (Virginia Tech) to support Sustainable Agriculture and Natural Resource Development for developing countries.
- Vivo. NAL began working with ARS to establish a semantic web application that enables the
 discovery of research and scholarship across the USDA. The application will allow better
 discovery for networking, collaboration and research. VIVO will also allow citizens to better
 discover USDA research. This application is called "VIVO" and is an Open Source, or free
 application jointly developed by Cornell University and the University of Florida. NAL will host
 VIVO for USDA.
- Interagency partnerships: NAL continued to be very active in developing and maintaining partnerships to provide digital information services. Nutrition.gov, invasivespeciesinfo.gov, science.gov, and worldwidescience.org are multi-agency and multi-national Web portals to which NAL contributes digital content and leadership. NAL also continued to participate actively in other interagency groups such as PHPartners (Public Health) and CENDI (scientific and technical information management) to promote and leverage NAL's work.